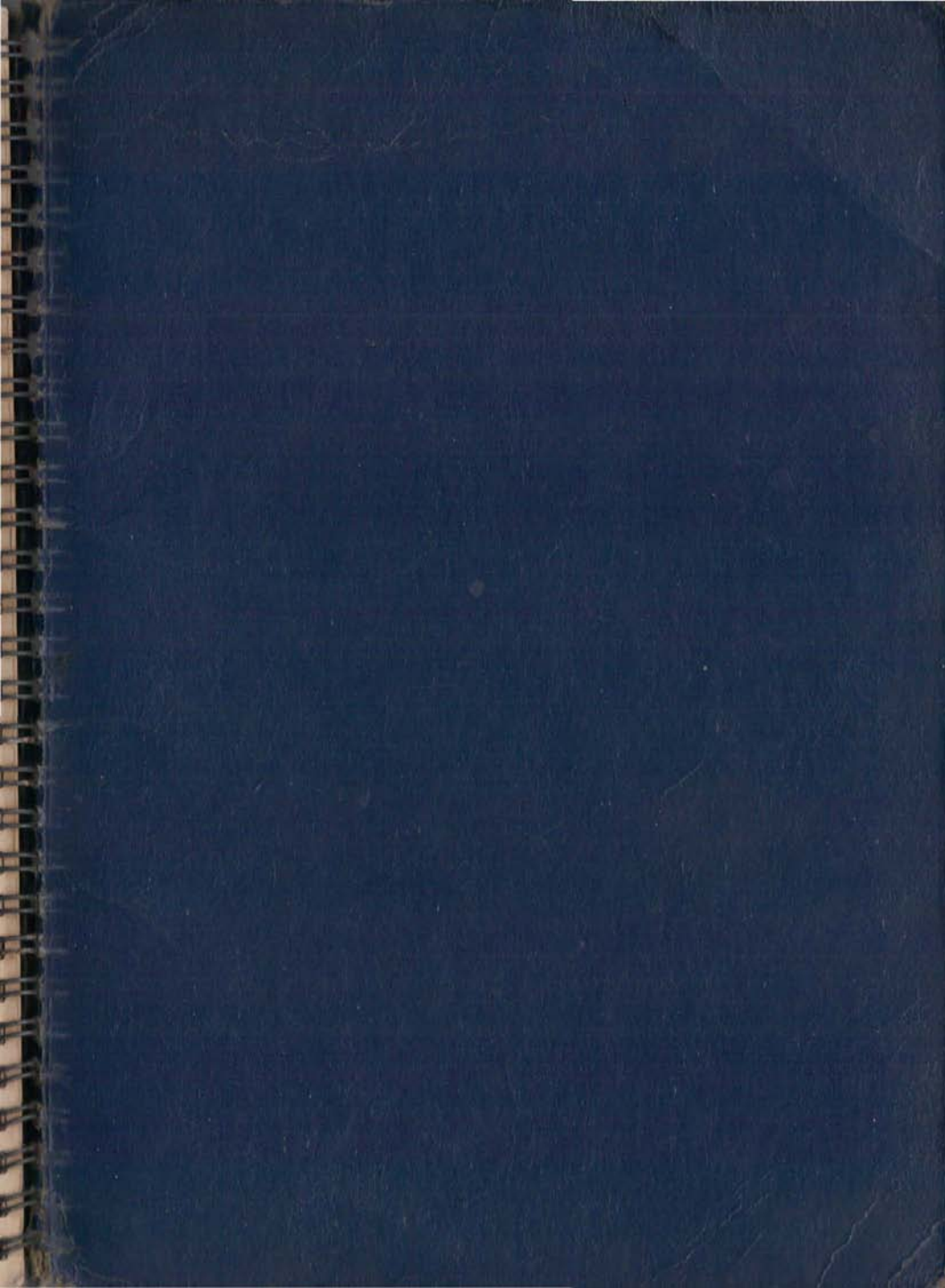


LABORATORY MANUAL OF CHEMISTRY

By JOSEPH I. ROUTH, Ph.D.

W. B. SAUNDERS COMPANY



Joyce Ann Farley
LABORATORY MANUAL OF
CHEMISTRY

Chapter 120.6
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album
Philadelphia and London

W. B. SAUNDERS COMPANY

1945

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PREFACE

The purpose of this Manual is to provide experiments that illustrate the material in the author's textbook, "Fundamentals of Inorganic, Organic, and Biological Chemistry." The experiments are arranged to follow the order of presentation in the book; however, they are sufficiently general in nature to be used with any standard textbook on the subject.

Although the arrangement of the Manual is similar to that of others in the field, the individual experiments have often been modified by use with several classes of nurses, and the selection of experiments has been governed by their usefulness to the student. In the author's experience, the best results in teaching have been obtained when the discussion of text material in lectures and recitations was followed closely by an opportunity to demonstrate the fundamental principles by suitable laboratory experiments.

The questions that accompany the exercises are designed to facilitate recording of the results of each experiment and to emphasize the fundamental principles involved. In many instances, the questions also cover practical applications of these principles to medicine and nursing. The answers to the questions may be obtained either from observations in the laboratory or from reference to the text.

An effort has been made to minimize the time spent by the students in such tedious procedures as the weighing of exact quantities of solids and the construction of laboratory apparatus. The amounts of solids are usually estimated and the preparation of pieces of apparatus such as delivery tubes, air condensers, and so forth, is left to the instructor.

The author is especially indebted to Dr. M. Heinrich for his assistance in the selection of the experiments included in the Manual. He also takes pleasure in acknowledging the valuable suggestions and criticisms of the material included in the experiments by all of the graduate students who have assisted him in teaching the course of chemistry to nurses. The illustrations of laboratory apparatus at the beginning of

the manual are reproduced through the courtesy of the Central Scientific Company of Chicago and the Arthur H. Thomas Company of Philadelphia.

JOSEPH I. ROUTH

IOWA CITY

June, 1945

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SUGGESTED APPARATUS FOR STUDENT DESKS

- 3 beakers, Pyrex, 100 cc., 250 cc., and 400 cc. capacity
- 2 bottles, 100 cc. capacity, to take a No. 5 or 6 stopper
- 1 Bunsen burner with rubber tubing; 1 wing tip for burner
- 1 evaporating dish, porcelain, 4 in. diameter
- 1 file, triangular, 5 in.
- 1 package filter paper, 12.5 cm. diameter, qualitative
- 2 flasks, Erlenmeyer, 125 cc. and 250 cc. capacity
- 1 funnel, $2\frac{1}{2}$ in. diameter
- 2 glass plates, 3 x 3 in.
- 2 graduated cylinders, 10 cc. and 50 cc. capacity
- 2 vials of litmus paper, 1 red and 1 blue
- 1 box matches
- 1 mortar and pestle, approximately 3 in. diameter
- 1 pipette, Mohr measuring type, 5 cc. capacity
- 1 ring stand, iron, with 1 burette clamp and 1 ring, iron, 3 in. diameter
- 2 stirring rods, glass, 6 in. long, 5 mm. diameter
- 10 test tubes, Pyrex, $\frac{3}{4}$ in. x 6 in.
- 1 test tube brush; 1 test tube holder; 1 test tube rack
- 1 thermometer, -10° to 110° C.
- 1 tongs, brass
- 1 towel
- 1 watch glass, 4 in. diameter
- 1 wire gauze

SUGGESTED APPARATUS FOR GENERAL USE

- Corks, assorted sizes
- Glass tubing, 6, 7, and 8 mm. diameter
- Graduated cylinders, large sizes
- Laboratory scales with sets of weights
- Medicine glasses, graduated
- Microscopes
- Milk bottles, quart
- Pneumatic troughs
- Rubber stoppers, sizes 3 to 7 with 1 and 2 holes.
- Rubber tubing, for connections
- Water tumblers



Fig. 1.—Beaker.



Fig. 2.—Gas-collecting bottle.



Fig. 3.—Reagent bottle.

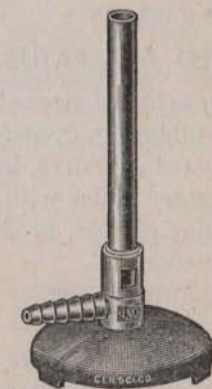
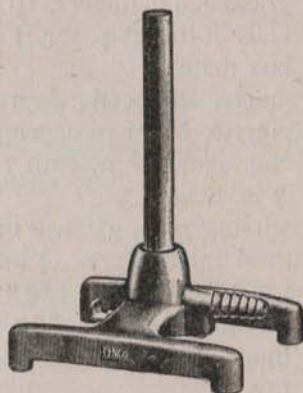


Fig. 4.—Bunsen burner.



Fig. 5.—Burette clamp.



Fig. 6.—Evaporating dish.



Fig. 7.—File, triangular.

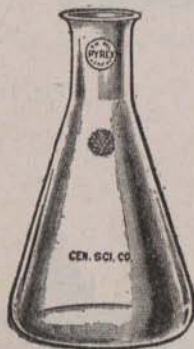


Fig. 8.—Flask, Erlenmeyer.

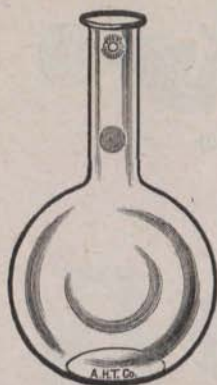


Fig. 9.—Flask, Florence.



Fig. 12.—Graduated cylinder.

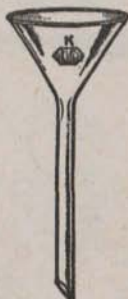


Fig. 10.—Funnel.



Fig. 13.—Mortar and pestle.



Fig. 11.—Funnel rack, to fasten to ring stand.

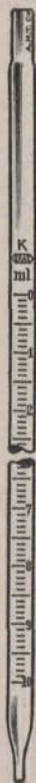


Fig. 14.—Pipette, Mohr measuring type.

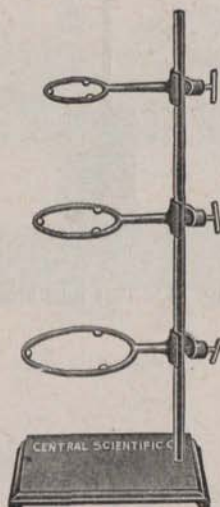


Fig. 15.—Ring stand, with rings.



Fig. 16.—Spoon, deflagrating.



Fig. 17.—Test tube.

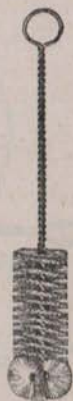


Fig. 18.—Test tube brush.

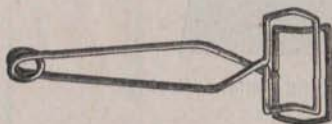


Fig. 19.—Test tube holder.

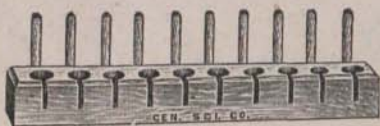


Fig. 20.—Test tube rack.

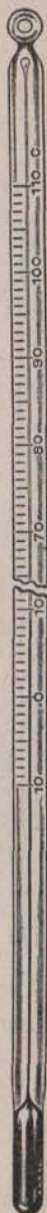


Fig. 21.—Thermometer.

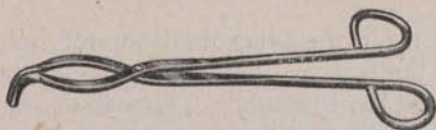


Fig. 22.—Tongs, brass.

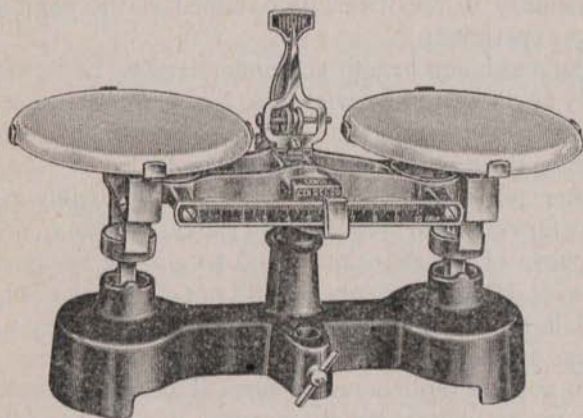


Fig. 23.—Trip scales.

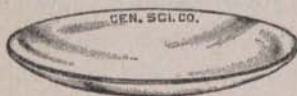


Fig. 24.—Watch glass.



Fig. 25.—Wing tip for burner.



Fig. 26.—Wire gauze, with asbestos center.

Study

SUGGESTIONS FOR THE STUDENT

Form the habit of reading the laboratory experiments and the section of the text that pertains to the laboratory material before coming to the laboratory. This will not only enable you to save time but will increase your understanding of the material covered by the experiments.

Record your results as you observe them. Do not rely on your memory to recall results obtained at the beginning of a lengthy experiment.

The maximum benefit and understanding of an experiment can only be obtained by performing it yourself. Always do every experiment that is assigned and record your own results before comparing them with those of other students.

After performing an experiment and possibly consulting the text for reference material, you should be able to understand the purpose of the experiment and to answer the questions in the manual. If certain points are still not clear, ask the instructor to explain them to you. Do not leave the laboratory until your questions are answered.

Do not use the laboratory manual as a cook book. Within reason, try to understand the purpose of each experiment and the reactions that are involved. Mere mechanical manipulation leads to a lack of understanding of the general principles of chemistry that are illustrated by the laboratory experiments.

GENERAL LABORATORY DIRECTIONS

Obtain a rubber apron or a laboratory coat to protect your clothing from acids or alkalis.

Provide yourself with a ruler that is graduated in inches and centimeters.

Always use clean apparatus for each experiment. At the end of the laboratory period clean all dirty equipment in preparation for the next period.

Read the labels on all bottles before removing the chemical for use in an experiment. Avoid wasting chemicals by taking only the amount that is required for the experiment. If you accidentally take more than you need, give it to another student; do not put it back in the reagent bottle or you may contaminate the entire contents.

While pouring from reagent bottles, hold the stopper between the first and second fingers of the right hand. Always replace each stopper before using another bottle to prevent interchange of stoppers and contamination of the contents of the bottles. When you pour strong acids or alkalis in the drain or sink, be sure to have plenty of water running from faucets to dilute the reagents and prevent them from corroding the pipes. If strong acids or alkalis are spilled on the skin or clothing they should be washed off immediately with large quantities of water. Any remaining acid may be neutralized with a dilute solution of ammonium hydroxide; alkalis may be neutralized with a dilute solution of acetic acid. The reagents should then be rinsed off with more water.

Never use inflammable liquids such as ether, benzene, gasoline, or alcohol near a flame.

If a fire occurs in a beaker or evaporating dish it can best be extinguished by covering the vessel with a wet towel. More extensive fires should be extinguished with sand, a blanket, or a chemical fire extinguisher.

USE OF THE BUNSEN BURNER

Examine a Bunsen burner. You will observe that the gas inlet is at the base of the burner. As the gas enters the burner

tube it is mixed with air that is admitted through holes near the base of the burner. The proper mixture of air and gas, when lighted, produces a non-luminous blue flame which is hottest at the tip of the inner blue cone. If insufficient air is mixed with the gas a yellow luminous flame will result which will deposit soot on any object that is placed in the flame. If too much air is mixed with the gas it will blow out the match when an attempt is made to light it. When lighting a burner allow the gas to flow a few seconds before applying a match. This will prevent the flame from being drawn back into the burner tube. If this occurs turn off the gas at once and start over. Adjust the air regulator on your burner until you have a pale blue non-luminous flame with a distinct inner cone.

CUTTING GLASS TUBING

Lay a piece of glass tubing on a flat surface and make a deep scratch across the tube with a triangular file. Then grasp the tube with both hands placing the thumbs on the opposite side of the tube from the file mark. With a slight pressure of the fingers and thumbs attempt to bend the tubing as if you were breaking a twig at the file mark. A little practice will determine the depth of cut and amount of pressure that is required for a clean break.

BENDING GLASS TUBING

After attaching the wing tip to the burner, light the flame. Hold a 10 to 12 inch length of glass tubing at each end and heat the central portion of the tube in the entire length of the wing-shaped flame. To insure even heating, rotate the tubing slowly, being careful not to twist it when it is softened by the flame. When the glass is softened to a point where the hot portion begins to sag, remove it from the flame and bend to the proper shape, holding it in position until the glass hardens. After a piece of glass tubing has been cut and bent to the desired shape the rough ends should be rotated in the flame until they become smooth. Tubes that are to be inserted into stoppers or rubber tubing or used as drinking tubes should have all ends heated, or fire polished, to prevent injury from the sharp edges.

To draw out a piece of tubing in the preparation of an eye

dropper or a pipette, the tube should be heated in the regular flame of a Bunsen burner. Slowly rotate the tube until the hot portion becomes soft, then remove from the flame and draw out the tube by pulling both hands apart.

HEATING OF LIQUIDS

A. IN A TEST TUBE: Hold the test tube with a test tube holder in a slanting position above the flame of a Bunsen burner.

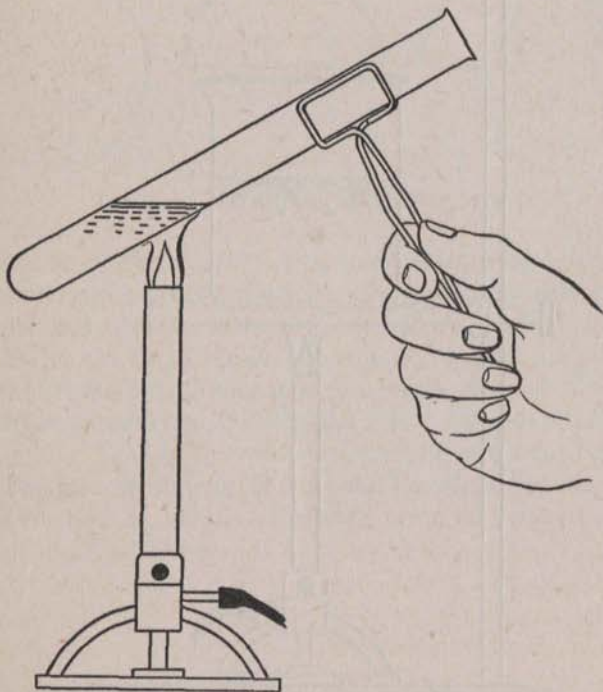


Fig. 27.—Heating of liquids in a test tube.

(Figure 27.) The test tube should not be heated directly on the bottom as this will cause the contents to spurt out of the tube. Heat the liquid near its surface in such a way that the liquid boils gently without boiling over, being careful not to heat the glass above the liquid as this will cause the tube to break. Always

12 General Laboratory Directions

point the mouth of the test tube away from anyone working near you when you heat its contents.

B. IN A BEAKER: The beaker and its contents should be supported about 2 to 3 inches above the Bunsen burner by an iron

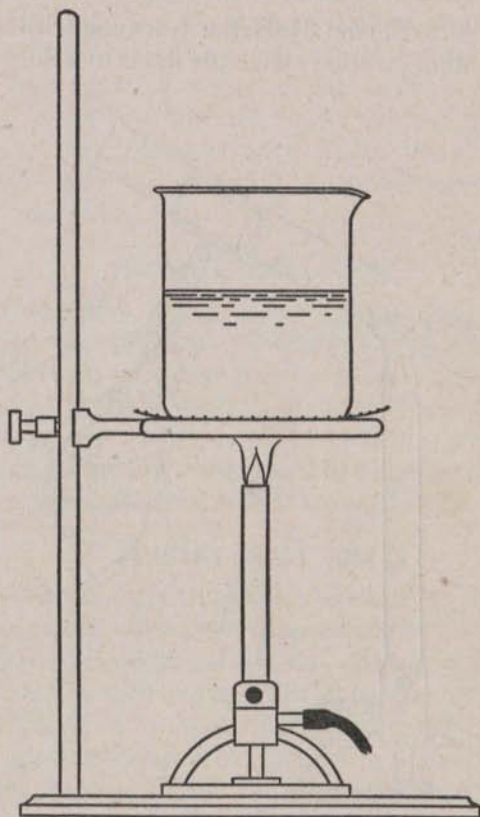


Fig. 28.—Heating of liquids in a beaker.

ring on a ring stand, or by a tripod. (Figure 28.) A square of wire gauze should always be placed between the beaker and the flame (supported by the ring or tripod) to prevent breakage of the glass by unequal heating.

FILTRATION

Fold a piece of filter paper in half, then in quarters. Open the folds to form a cone with three thicknesses of paper on one side and one on the other. (Figure 29.) Place the cone in a glass funnel, fitting it snugly to the sides by making a new crease in the paper if necessary. While the paper is held in position with the fingers, moisten it with distilled water to hold it in place.

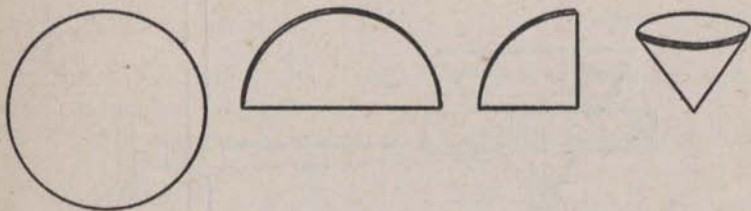


Fig. 29. —Method of folding filter paper.

To remove a suspended solid or precipitate from a solution by filtration, first support the funnel, containing the filter paper, on a funnel rack or in an iron ring; then place a beaker under the funnel with the tip of the funnel pressing against the side of the beaker (Figure 30); finally pour the solution to be filtered into the funnel carefully to prevent the liquid from overflowing the filter paper. This is best done by pouring the solution down a glass rod into the funnel. The filtered liquid, or filtrate, will collect in the beaker, whereas the solid material will remain in the cone of the filter paper.

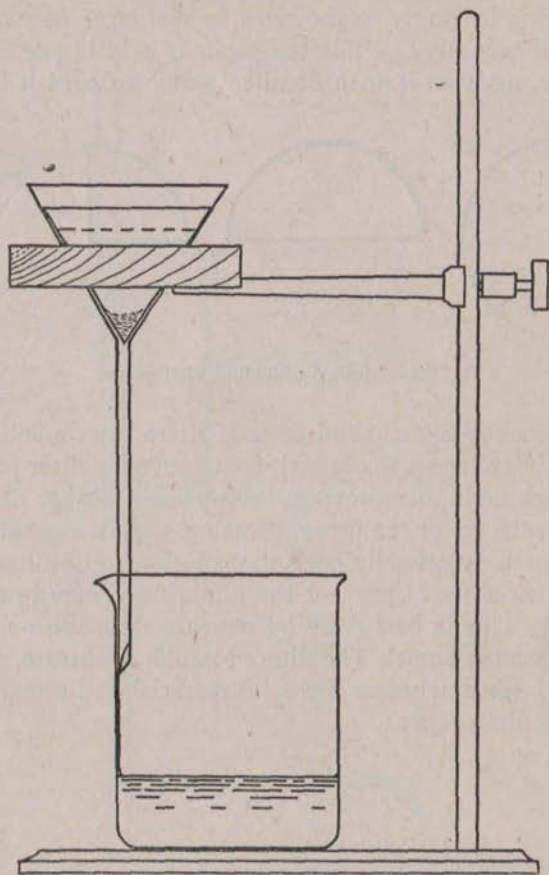


Fig. 30.—The filtering process.

THE METRIC SYSTEM

SPECIAL REQUIREMENTS:¹ Meter sticks, graduated cylinders, 250 cc. and 1000 cc. sizes, medicine glasses, water tumblers, quart milk bottles, scale graduated in pounds, laboratory balances and sets of weights, crushed ice.

Experiment 1. Linear Measurements: Measure your height on a scale graduated in inches. 65 in. Convert this value to centimeters 162.5 cm. With a ruler divided in inches and centimeters make the following measurements:

Your span (tip of thumb to tip of little finger with hand spread) is 8 in. 20 cm.

The width of your thumbnail is _____ mm.

The length of a test tube is 15 cm. 7 in.

The length of the middle joint of your middle finger is 2.5 cm. 1 in.

The diameter of the top of one of your funnels is 70 mm. 7 cm. 2 1/2 in.

A six inch length of your ruler is equivalent to 15 cm.

A distance of one inch on your ruler is equivalent to 25 mm.

Experiment 2. Volume: With graduated cylinders measure the volume of several types of containers.

¹ In general, the special requirements of each section are listed in the order they appear in the experiments in that section. When a solution is not specified the pure chemical compound is indicated. In many cases the exact concentration of a solution is not mentioned in the experiment, however the concentrations of solutions are always given in the special requirements section.

Directions for the preparation of special reagents will be found in footnotes following the experiment in which they are used. These footnotes are referred to after the name of the reagent in the section of special requirements.

A test tube contains 25 cc.

A medicine glass filled to the teaspoon mark contains 3 cc.

When filled to the tablespoon mark 14 cc.

When filled to the one ounce mark 26 cc.

A water tumbler contains _____ cc.

A quart milk bottle has a capacity of _____ cc.

A standard 8 ounce measuring cup contains a volume of water equivalent to _____ cc.

Experiment 3. Weight: Weigh yourself on a large scale calibrated in pounds. 114 lb. Express your weight in kilograms. 51.8 kg.

Weigh a 50 cc. graduated cylinder on a laboratory balance: weight _____ gm.

With exactly 25 cc. of water in the cylinder it weighs _____ gm.

Weight of 25 cc. of water 25 gm.

Weight of 1 cc. of water 1 gm.

A 1-pound weight is equivalent to _____ gm.

Experiment 4. Temperature: The temperature in the laboratory is 26 ° C. This is equal to _____ ° F.

The temperature of the tap water is _____ ° C. or _____ ° F.

Fill a beaker about half full of distilled water and heat to boiling.

The temperature of boiling water is 100 ° C. or 212 ° F.

Fill another beaker about half full of crushed ice and carefully

insert a thermometer to the bottom of the beaker, after a few minutes remove the thermometer and rapidly read the temperature.

The temperature is 0 ° C. or 32 ° F.
Dry ice has a temperature of -80° C. What would this be
on the Fahrenheit scale? ° F.

Body temperature is normally 98.6° F. or 37 ° C.

PHYSICAL AND CHEMICAL CHANGES

SPECIAL REQUIREMENTS: Platinum wire sealed in glass tubes, copper wire, ice and salt mixture, wooden splints, 2 per cent solution of silver nitrate.

Experiment 5. Effect of Heat on Metals: Heat the end of a platinum wire in the flame of a burner.

Do you observe any changes on heating? yes

After the wire has cooled are any changes from the original properties apparent? no

Is this a physical or a chemical change? physical
Heat the end of a piece of bright copper wire in the flame. (The wire may be held with tongs or test tube holder.)

Does a change occur on heating? yes

Describe the change. The wire becomes dark colored

Allow the wire to cool. Does the copper still retain its original properties? no

Has a new substance been formed? yes

Compare the properties of the coating on the heated portion of the wire with the original copper. After heated

the coating is darker, and has less lustre

Is this a physical or chemical change? physical

Experiment 6. Changes in Water: What are the outstanding physical properties of water? colorless, tasteless, odorless, and liquid

Heat a few cc.'s of water to boiling in a test tube. While the water is boiling hold a small flask of cold water above the mouth of the test tube.

What forms on the outside of the flask? Condensation

Into what form was the water changed during boiling?

water vapor

Would you consider this change from a liquid to a gaseous state a physical or chemical change? physical

What observation helps substantiate this conclusion?

The water first changes into steam then condenses as water

Freeze a few cc.'s of water in a test tube by immersing the tube in a mixture of ice and salt.

Compare the properties of the frozen water with those of the original liquid. the frozen is a solid, and

the water is a liquid

Allow the ice to melt. What is formed? water

Is the freezing of water a physical or chemical change?

physical

What are your reasons for this answer? Because it can

be changed back into the original state

Experiment 7. Burning of Wood: Place a few small pieces of wood in a test tube and heat until the material in the tube becomes charred.

Has a new substance been formed? yes

Compare the properties of the new substance with those of the original wood. The new is very dark, while

the old was yellow

Has a physical or chemical change taken place?

Chemical

Explain your reasoning. the wood just has changed to wood charcoal. The physical properties have undergone a change

Experiment 8. Changes Caused by Light: Moisten a small spot in the center of each of two pieces of filter paper with a dilute solution of silver nitrate. Be careful not to spill the solution on your skin as it will cause a stain; if accidentally spilled on the skin wash it off immediately with large quantities of water. Cover one piece of paper or place it in a dark place. The other piece should be exposed to sunlight for 30 minutes. At the end of this time examine both pieces of filter paper.

Can you observe any difference between them? The one

in the dark remains unchanged & the
one in the light circles.

Has the light caused a change to take place? yes

Has a new substance been formed from silver nitrate?

yes

What type of change is this? chemical

ELEMENTS, COMPOUNDS, AND MIXTURES

SPECIAL REQUIREMENTS: Elements listed in Experiment 9, magnesium ribbon, cane sugar, white sand, sodium chloride.

Experiment 9. Properties of Elements: Examine samples of the following elements listing their outstanding properties (physical state, color, odor, luster, texture, metal, or non metal etc.) in the table.

ELEMENTS

PROPERTIES

Iron	heavy, metal, dark color, is hard
Copper	soft copper colored metal.
Sulfur	non metallic, strong odor, yellow in color
Aluminum	light metal, has luster and is light
Mercury	heavy liquid, silver, white metal
Zinc	a bluish-white metal. Can be exposed.
Iodine	an element with sharp odor and taste
Lead	soft bright metal

Experiment 10. Formation of Compounds from Elements: Describe the properties of the element magnesium. has luster
and is very soft metal

This element will readily combine with oxygen to form the compound magnesium oxide. Place a short piece of magnesium ribbon in an evaporating dish and touch it with the flame of a burner.

What takes place in the dish? It burns and makes a
white bright light. It burns rapidly

Compare the properties of the new substance with those of the original element. The new substance is a
white powder unlike the old one
was a soft metal

Where did the oxygen in the new substance come from?

It came from the air

Is the new substance an element, a mixture or a compound?

Compound

Explain your reasons for this answer. Because the

oxygen & the magnesium can't be separated.

Experiment 11. Decomposition of a Compound into its Elements: Heat a small amount (enough to fill the rounded portion of a test tube) of sucrose in a test tube and observe the changes that take place.

What condenses on the walls of the upper portion of the test tube? gas

What elements are present in this substance? water
and oxygen

Heat until no further change occurs, allow the tube to cool, break it in a mortar and examine the contents.

Compare the properties of this material with those of

sucrose. sugar was a white crystal and the new substance was brown with a distinct odor and in a more solid state

What is this residual material? Carbon

Of what elements is sucrose composed? Carbon, hydrogen
and oxygen

Experiment 12. Properties of Mixtures: Examine small amounts of white sand and sodium chloride (table salt). List their outstanding properties:

Sand coarser texture, dark color, lustrous

Sodium chloride fine texture

Mix equal quantities of each substance in a dry mortar and grind them with a pestle until a fine white powder is formed.

Does this powder appear to be homogeneous? yes

Has an apparent chemical change taken place? no

Transfer a portion of the white powder to a beaker and add 100 cc. of water to dissolve the salt. Filter through filter paper, washing the residue from the beaker with fresh portions of water.

After tasting the solution (filtrate) and the residue on the filter paper would you conclude that sodium chloride can be separated from sand by physical or mechanical means?

physical
Has a chemical change occurred during this process?

no

How would you differentiate between compounds and mixtures?

Compounds have undergone a chemical change and mixtures have not.

OXYGEN, OXIDATION AND REDUCTION

SPECIAL REQUIREMENTS: Potassium chlorate, manganese dioxide, bent glass delivery tube in a one-hole rubber stopper (Figure 31), pneumatic trough, wooden splints, limewater, powdered sulfur, steel wool, 3 per cent solution of hydrogen peroxide, Javelle water (footnote p. 27), sodium thiosulfate, oxalic acid crystals, white cotton cloth stained with materials in Experiment 17 and cut in small squares.

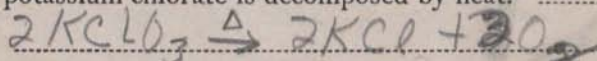
OXYGEN

Experiment 13. Preparation of Oxygen: (This experiment should be performed in pairs.) Mix thoroughly 10 gm. of potassium chlorate, KClO_3 , and 1 gm. of manganese dioxide, MnO_2 , in an Erlenmeyer flask. Insert in the flask a rubber stopper connected to a bent glass delivery tube. Clamp the flask to a ring stand in such a position that the end of the delivery tube extends into a pneumatic trough under the rack that supports the collecting bottles (Figure 31). Heat the flask gently and allow the first bubbles to escape under water to remove the air from the flask and delivery tube. Then place a collecting bottle, completely filled with water, over the opening of the delivery tube and fill it with oxygen by displacing the water it contains. To remove a bottle of gas, cover the mouth with a glass square, lift out of the water and place the bottle right side up on the desk. Do not stop heating the flask until the delivery tube has been removed from the water to prevent the sucking back of water into the hot flask. Collect three bottles of oxygen for use in the following experiment.

Why are the bottles kept covered with a glass plate?

*So the oxygen would not become
aminated*

Write an equation showing how oxygen is formed when potassium chlorate is decomposed by heat.



What is the purpose of the manganese dioxide?

To increase the formation of oxygen.

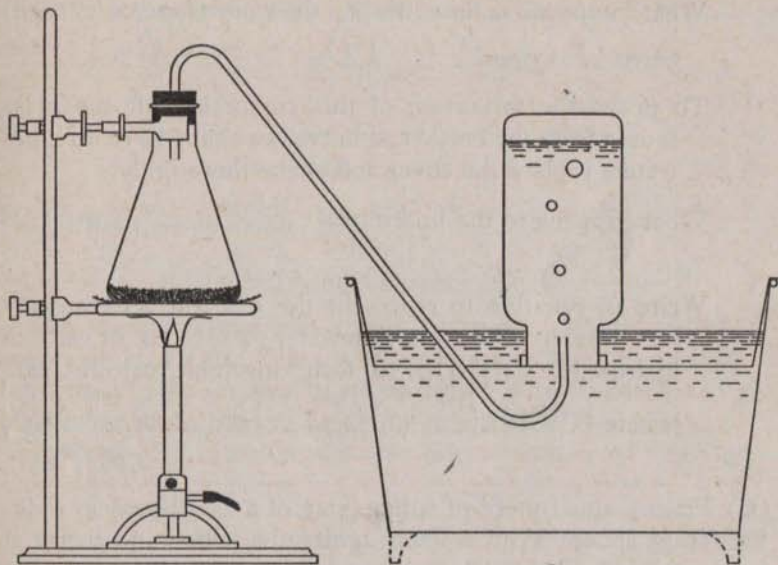


Fig. 31.—Apparatus for the preparation of oxygen.

*Experiment 14. Properties of Oxygen:*¹ (A) After an examination of one of the bottles of oxygen describe such properties as its physical state, odor, taste and color.

Description of properties. *It was an odorless, tasteless, and rather cloudy gas.*

(B) Place a small piece of wood in a deflagrating spoon and set it on fire with the flame of a burner. After it has burned awhile, blow out the flame and quickly thrust the glowing splinter into a bottle of oxygen, covering the opening with the glass square.

Compare the burning of the wood in air and in oxygen.

It burns much brighter & faster in oxygen.

¹ If insufficient time is available for students to perform the experiment on the preparation of oxygen, its properties may be studied by supplying the gas from a commercial cylinder of oxygen.

What compound is formed when the wood charcoal (carbon) burns in oxygen? $C + O_2 \rightarrow CO_2$

To prove the formation of this compound lift the glass square from the bottle and introduce about 10 cc. of limewater; replace the cover and shake thoroughly.

What happens to the limewater? *It becomes a white milky color.*

Write an equation to represent the reaction between carbon dioxide (CO_2) and limewater (a solution of calcium hydroxide, $Ca(OH)_2$) to form insoluble calcium carbonate ($CaCO_3$). $Ca(OH)_2 + CO_2 \rightarrow CaCO_3 + H_2O$

(C) Place a small piece of sulfur (size of a small pea) in a deflagrating spoon. With a flame ignite the sulfur and thrust it into another bottle of oxygen.

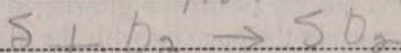
Describe the action. *First melts then burns with pale blue flame.*

When burning ceases, remove the spoon, immediately add a few cc.'s of water to the bottle, cover and shake thoroughly. Test the water with a piece of blue litmus paper

Result? *It turned red*

What gas was formed when sulfur combined with oxygen in the burning process? SO_2

Write an equation for the reaction of sulfur dioxide (SO_2) and water to form the acidic substance that turned the blue litmus red. $H_2O + SO_2 \rightarrow H_2SO_3$



(D) Add about 10 cc. of water to the third bottle of oxygen then immediately replace the cover on the bottle. (The water is to prevent the bottle from breaking.) Heat a small wad of iron wool, held by tongs, to redness in a flame. Quickly thrust the red hot material into the bottle of oxygen.

What happens? *It burns very rapidly in a bright, quick light flame.*

Two oxides of iron (ferric oxide Fe_2O_3 and ferrous oxide FeO) are formed when iron burns in oxygen. Write an equation to represent the union of iron and oxygen to

form ferric oxide. $\text{Fe}^{++} + \text{O}^{--} \rightarrow \text{Fe}_2\text{O}_3$

OXIDATION AND REDUCTION

Experiment 15. Oxidizing Agents: Examine a sample of hydrogen peroxide (H_2O_2). This is a common oxidizing agent that is often used in the removal of stains. When it decomposes it gives off oxygen to be used in oxidation reactions.

Write the equation for the decomposition of hydrogen peroxide. $\text{H}_2\text{O}_2 \rightarrow \text{H}_2 + \text{O}_2$

Javelle water¹ is a solution of sodium hypochlorite (NaOCl). This compound is an oxidizing agent often used in the bleaching of stains. Such commercial products as Chlorox and Hilex depend for their action on sodium hypochlorite.

From an inspection of the formula why would you suspect this compound possessed the properties of an oxidizing

agent? *Because it has a free oxygen atom to be given off.*

Experiment 16. Reducing Agents: Prepare a solution of sodium thiosulfate ($\text{Na}_2\text{S}_2\text{O}_3$) by dissolving approximately 5 gm. of the compound in 50 cc. of water. This substance is a reducing agent and is often termed "hypo" when used in photography and X-ray laboratories. Also prepare a saturated solution of oxalic acid $(\text{COOH})_2$ by dissolving as many crystals as possible in 50 cc. of warm water. This reducing agent is sometimes used to remove rust and iron stains from porcelain ware.

¹ Dissolve 10 gm. of chlorinated lime (bleaching powder) in 100 cc. of a 20 per cent sodium carbonate solution. Filter the mixture to remove the calcium carbonate and keep in well stoppered bottles.

What is a reducing agent?

It is an agent which causes reduction to become

Experiment 17. Stain Removal: Use the above oxidizing and reducing agents in an attempt to bleach pieces of cotton cloth previously treated with the stains listed in the following table. For best results small portions of the stained cloth should be cut off and soaked in the various bleaching agents. Make a record in the table of the agent that best removes each stain.

Stains	BLEACHING AGENTS			
	Oxidizing Agents		Reducing Agents	
	Hydrogen peroxide solution	Javelle water	Sodium thiosulfate solution	Saturated oxalic acid solution
Iodine	no	yes	yes	no
Ink	yes	yes	yes	yes
Silver nitrate	no	yes	no	no
Fruit juice	yes	yes	yes	yes
Potassium permanganate	no	no	no	yes
Blood				
Tobacco				
Iron rust	no	no	no	yes

WATER

SPECIAL REQUIREMENTS: Powdered citric acid, sodium bicarbonate, sodium sulfate, calcium chloride, copper sulfate crystals, plaster of paris, squares of waxed paper, potassium permanganate, 10 per cent solution of ammonium hydroxide, bent glass delivery tube in a one-hole rubber stopper (Figure 32), temporary hard water (footnote p. 33), soap solution, concentrated solution of sodium carbonate.

PROPERTIES

Experiment 18. Water as an Agent Necessary for Reaction: Mix together in a dry beaker a small amount (approximately 1 gm.) of powdered citric acid and an equal quantity of sodium bicarbonate (baking soda).

Do you observe any chemical reaction? no

Add 50-75 cc. of water to the beaker and stir. What happens?

It bubbles and fizzes

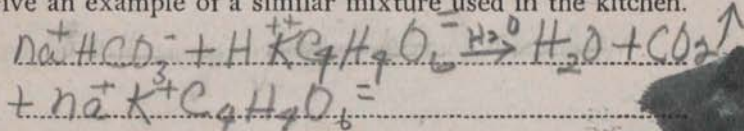
What is the nature of the gas formed by the action of acids

on bicarbonates?

How did the water function in this reaction? It is what

caused the reaction

Give an example of a similar mixture used in the kitchen.



Describe its action.

Experiment 19. Efflorescence and Deliquescence: Put a crystal of sodium sulfate in a dry evaporating dish and a granule of calcium chloride in a dry watch glass. Set them in your desk so they will be exposed to the air until the next period.

What changes occur in the sodium sulfate? Lost its water and became a dry powder

In the calcium chloride? It took up so much water it dissolved itself

Differentiate between efflorescence and deliquescence.

efflorescent is substance which give up water. Deliquescence takes up water of crystallization

How should efflorescent or deliquescent chemicals be stored to prevent changes? In an air tight container

Experiment 20. Water of Crystallization: Drop a small clear crystal of copper sulfate into a test tube and heat it gently with a Bunsen burner.

What substance condenses on the walls of the upper portion of the test tube? H₂O

Where did this water come from? The copper sulfate

What color is the copper sulfate after it is heated? white

Write the formula for anhydrous copper sulfate. CuSO₄

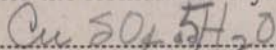
After the contents of the tube have cooled add a few drops of water.

Is there any change in the color of the copper sulfate?

It turned back to its original color

Explain this behavior. The water which had been taken out is now added giving it back its blue color

Write the formula for crystalline (hydrated) copper sulfate.



From the results of this experiment describe the difference

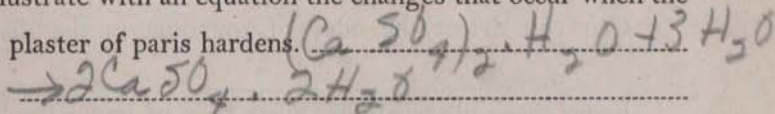
between hydrates and anhydrous compounds. Hydrates contain H_2O while anhydrous is free from water.

Experiment 21. Plaster of Paris: Place a small amount of plaster of paris on a piece of waxed paper. Carefully add water and mix until a soft pliable mass results. Mold this material around the bottom of a test tube or the end of a stopper. After a half hour remove the cast and examine the impression of the tube or stopper. The crystalline structure of the solid material can be observed by breaking off a piece of the cast.

Why are the above properties of plaster of paris important in the practice of medicine?

It is tough, it can be molded into a cast for surgical use.

Illustrate with an equation the changes that occur when the plaster of paris hardens.



PURIFICATION

Experiment 22. Distillation: (This experiment should be performed with a partner.) Place 50–75 cc. of water, a crystal of potassium permanganate, KMnO_4 , and a few drops of ammonium hydroxide in an Erlenmeyer flask. Insert in the flask a rubber stopper connected to a bent glass delivery tube as shown in Figure 32. The end of the tube should extend into a test tube placed in a beaker of cold water. Boil the liquid in the flask until about 5 cc. of fluid, called the distillate, has condensed in the test tube.

How does the color of the distillate compare with the liquid in the Erlenmeyer flask?

The water in the flask was deep purple in color + the distillate was very clear.

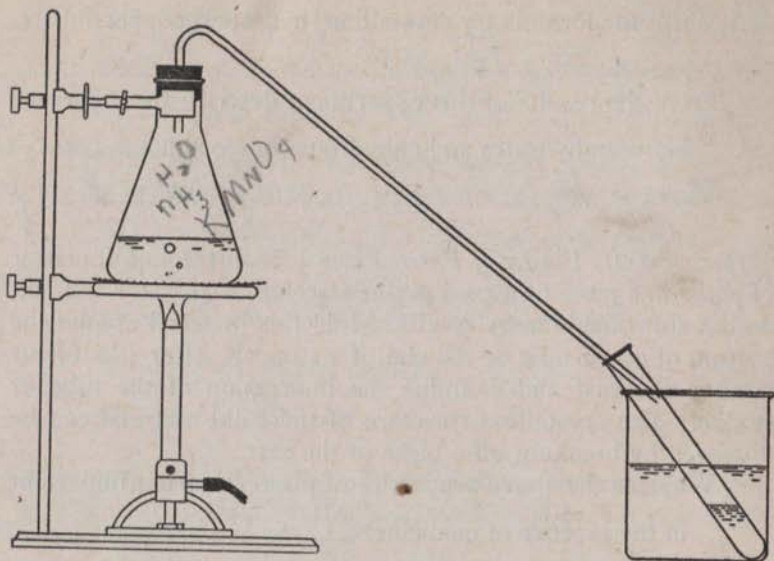


Fig. 32. — Apparatus for the distillation of water.

Can you detect the odor of ammonia in the distillate?

yes

From these observations, what type of impurities can be removed from water by distillation?

The impurities that will boil

What type of impurities cannot be removed by distillation?

those which will not boil

Will distillation remove any other types of impurities from water?

yes, any volatile

Explain

any non-volatile can't be removed by distillation

SOFTENING OF HARD WATER

Experiment 23. Temporary Hard Water: To 5 cc. of temporary hard water¹ add soap solution a few drops at a time, shaking after each addition, until suds are readily formed. Repeat the test using 5 cc. of distilled water instead of the temporary hard water.

Which water formed suds the most readily? 11 drops distilled H₂O

The least readily? Temporary hard water

Boil a sample of temporary hard water for about one minute.

What happens? CaCO₃ come out

Filter and test 5 cc. of the filtrate with soap solution as before.

Did boiling alter the hardness of the temporary hard water? Yes

Why is water, containing bicarbonates of calcium, magnesium, and iron, called temporary hard water? because they can be removed by heating

Write an equation to illustrate the softening of temporary

hard water by boiling. $\text{Ca}(\text{HCO}_3)_2 + \Delta \rightarrow \text{CaCO}_3 \downarrow + \text{H}_2\text{O} + \text{CO}_2$

Experiment 24. Permanent Hard Water: Test 5 cc. of permanent hard water (distilled water plus magnesium sulfate) for hardness with soap solution.

How do the results compare with those for distilled water?

CaCO₃ not removed

For temporary hard water? CaCO₃ is removed

Boil a sample of permanent hard water.

Does a precipitate form? no

Test a 5 cc. portion for hardness with soap solution as before.

¹ Prepared by passing carbon dioxide gas through limewater until the precipitate that forms has redissolved.

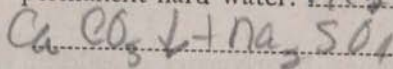
Has the water been softened by boiling? no

To another 5 cc. portion of permanent hard water add a concentrated solution of sodium carbonate (washing soda), dropwise with stirring until a precipitate no longer forms on further addition of the solution. Filter and test the filtrate for hardness with soap solution.

Has the addition of sodium carbonate softened the permanent hard water? yes

Name several other compounds that can be used to soften permanent hard water. _____

Write an equation to show how sodium carbonate softens permanent hard water. $\text{Na}_2\text{CO}_3 + \text{CaSO}_4 \rightarrow$



Take 5 cc of hard water and 0

SOLUTIONS

SPECIAL REQUIREMENTS: Alcohol, iodine crystals, powdered boric acid, sodium chloride, bottles of carbonated beverage, copper sulfate crystals, limewater, 1 per cent starch solution, lamp system throwing a strong beam of light, 0.1 per cent solution of bile salts, Cellophane tubing 1 to 2 in. wide, 10 per cent sodium sulfate solution, 5 per cent solution of barium chloride, iodine solution (footnote p. 63), blood, microscopes, slides, and cover slips.

FACTORS AFFECTING SOLUBILITY

Experiment 25. Influence of the Solvent: Place 5 cc. of water in one test tube and 5 cc. of alcohol in another. Add a small crystal of iodine to each tube. Mix well.

In which solvent is the iodine soluble? alcohol

What is tincture of iodine? iodine mixed with alcohol

Experiment 26. Influence of the Solute: Place a small quantity of powdered boric acid in one test tube (enough to fill the rounded portion of the tube) and an equal quantity of sodium chloride in another. Add water 1 cc. at a time to each tube, shaking the mixture after each addition. Record the number of cc.'s required to completely dissolve each compound.

The boric acid dissolved in 18 cc.'s.

The sodium chloride dissolved in 3 cc.'s.

Which solute is more soluble in water? sodium chloride

Experiment 27. Influence of Temperature: (A) Solubility of Gases: Open a cold bottle of carbonated beverage and pour some of the liquid into a beaker.

What are the bubbles that are continually escaping from the solution? carbon dioxide

Carefully open another bottle which has stood at room temperature (75°-80° F.). Pour some of the contents into a beaker.

How does the rate of evolution of bubbles compare with that of the cold liquid? Not as many

Are gases more soluble or less soluble at higher temperatures? less soluble

In what way does pressure affect the solubility of gases?

It increases it

(B) Solubility of Solids—Pulverize a large crystal of copper sulfate in a mortar. Place 5 cc. of water in each of two test tubes. Heat the contents of one tube to boiling, then add equal quantities of the powdered copper sulfate to each tube.

Is copper sulfate more soluble at high or at low temperatures? High

Heat 5 cc. of a saturated solution of calcium hydroxide, $\text{Ca}(\text{OH})_2$, (limewater) until it starts to boil. Compare the hot solution with 5 cc. of a similar calcium hydroxide solution that has not been heated.

What difference is apparent? Precipitate in hot water

Is calcium hydroxide more soluble or less soluble at high temperatures? less soluble

How does this behavior compare with that of copper sulfate? Copper sulfate is more soluble in high temp.

With other solids? Most other solids are more soluble

With gases? Gases are less soluble at high temp.

PREPARATION OF SOLUTIONS

Experiment 28. Preparation of Saturated and Percentage Solutions: Weigh 10 gm. of boric acid crystals on a piece of paper and transfer to a clean mortar. Grind to a fine powder and gradually add small portions of the solid to 100 cc. of distilled water (in an Erlenmeyer flask) shaking after each addition, until a slight excess remains. Weigh the remainder of the crystals.

How many grams of boric acid were required to prepare

100 cc. of a saturated solution? 2 1/2What is the per cent concentration of the solution? 4 per cent

Prepare 100 cc. of a 10 per cent sodium chloride solution. The group (5 to 6 students) in each row of desks should prepare 500 cc. of physiological salt solution (0.9 per cent). Save these solutions for use in Experiments 29 and 32.

COLLOIDAL SOLUTIONS

Experiment 29. Properties of Colloidal Solutions: Fill a test tube half full of 1 per cent starch solution.

Is the solution clear? no, it is milkyAre any solid particles visible in the solution? no

Filter the solution into another test tube.

Does filtration change the appearance of the solution?

no

Pass a strong beam of light through the starch solution in the test tube. Repeat, using a test tube containing a solution of sodium chloride.

Can this phenomenon, called the Tyndall Effect, be used to distinguish between a colloidal solution (starch) and a true solution (sodium chloride)? yes

Explain your reasoning.

Starch solution is smooth & the NaCl solution has tiny particles mixed with it.

PROPERTIES OF SOLUTIONS

Experiment 30. Surface Tension: Place about 5 cc. of water in one test tube and an equal quantity of 0.1 per cent bile salt solution in another. Place the tubes in a test tube rack and carefully sprinkle a small amount of powdered sulfur on the surface of each liquid.

Does the sulfur float on the water? yesOn the bile salt solution? no

What property of water is responsible for the sulfur floating on its surface? The high surface tension

Has the surface tension of water been altered by the presence of bile salts? yes

Which liquid has the highest surface tension? water

Why? just a property of H_2O

Experiment 31. Dialysis: Thoroughly wet a piece of Cellophane tubing about 6 inches long that has holes punched in it about $\frac{1}{2}$ inch from each end. Mix equal quantities (about 20 cc. of each)

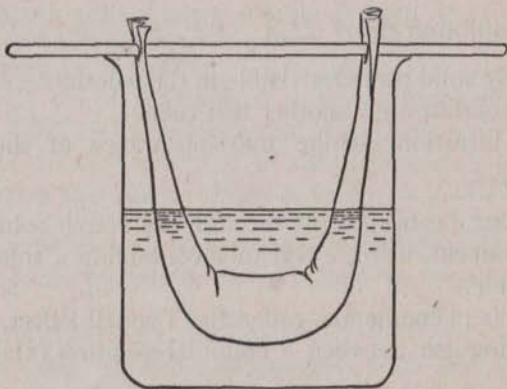


Fig. 33.—The dialysis of a solution.

of a 1 per cent starch solution and a 10 per cent sodium sulfate (Na_2SO_4) solution in a beaker. Pour this solution into the cellophane tubing that has been bent to form a U tube until the lower half of the U is filled. Suspend the U tube in a 250 cc. beaker by passing a stirring rod through the holes in each end and resting the rod on the top of the beaker (Figure 33). Add distilled water to the beaker until it reaches the level of the liquid in the U tube. After about one half hour remove the U tube and test a portion of the liquid in the beaker with a few drops of a barium chloride solution.

What do you observe? _____

If the sodium sulfate passed through the Cellophane membrane a white precipitate of barium sulfate will be formed. Test a second portion of the liquid in the beaker with dilute iodine solution (a blue color is a positive test for starch).

Results?

Why is the process of dialysis often used to separate particles in true solutions from those in colloidal solutions?

.....

.....

Give examples of other membranes that could be used as dialyzing membranes.

.....

Experiment 32. Osmosis: Arrange a series of test tubes as follows:

No. 1 contains 4 drops of blood and 5 cc. of distilled water.

No. 2 contains 4 drops of blood and 5 cc. of physiological salt solution (prepared in Experiment 28).

No. 3 contains 4 drops of blood and 5 cc. of 10 per cent sodium chloride solution (from Experiment 28).

Shake the tubes and allow to stand a few minutes.

Examine the contents of the three tubes, noting any difference in color or transparency.

.....

Place a drop of the mixture from each tube on a glass slide and examine it under a microscope.

Which solution did not affect the size or shape of the blood cells?

In which solution did the red blood cells burst and lose their contents?

Which solution caused the cells to shrink and lose their spherical shape?

burst

What is hemolysis? *splitting apart of b.c.*

What is crenation? *shrinking of cells,*

Why did osmosis occur in tubes No. 1 and 3?

In which direction does water flow through the membrane
of the red blood cell in hemolysis?

In crenation?

ACIDS, BASES, AND SALTS

SPECIAL REQUIREMENTS: Powdered sulfur, deflagrating spoons, 10 per cent solutions of hydrochloric, nitric, sulfuric, and acetic acids, phenolphthalein solution (footnote p. 42), granulated zinc, sodium carbonate, sodium bicarbonate, calcium oxide (unslaked lime), 10 per cent solutions of sodium hydroxide and ammonium hydroxide, limewater, 2 per cent silver nitrate solution, 5 per cent solutions of sodium sulfate, barium chloride, sodium carbonate, sodium chloride, ammonium chloride and copper sulfate, 2 per cent solution of sodium borate.

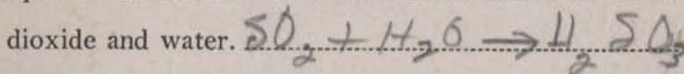
ACIDS

Experiment 33. Preparation of Sulfurous Acid: Place a small quantity of powdered sulfur in a deflagrating spoon. Ignite the sulfur in a flame and lower the spoon into a bottle that contains a layer of water approximately one-half inch in depth. Allow the sulfur to burn for 1 to 2 minutes then immerse the contents of the spoon into the water. Remove the spoon. Cover the bottle and thoroughly mix the solution. Place a drop of the solution on a strip of blue litmus paper.

What happens? *It turns it red*

When sulfur burns it forms sulfur dioxide gas. Write the equation for the formation of sulfurous acid from sulfur

dioxide and water.



Experiment 34. Properties of Acids: (A) Place in your rack 4 clean test tubes, each containing about 10 cc. of water. To the first tube add 2 or 3 drops of hydrochloric acid (HCl); to the second tube, 2 or 3 drops of nitric acid (HNO₃); to the third sulfuric acid (H₂SO₄); and to the fourth, acetic acid (HC₂H₃O₂). (All acids should be 10 per cent solutions.) Mix each by shaking, then using a clean glass rod, taste each solution.

Taste. *Sour*

Name some common foods which also have this taste.

Vinegar, salad dressings etc.

- (B) Transfer a drop of each solution, with the rod, to a piece of red and of blue litmus paper.

What happens? *Changes blue to red. Has no effect on the red*

- (C) Add one drop of phenolphthalein solution¹ to each tube.

Color? *White milky*

- (D) Add a small piece of zinc to a test tube containing 1 cc. of hydrochloric acid.

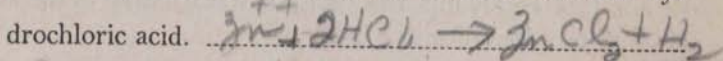
What happens when the test tube is gently warmed?

Bubbles of H₂ are produced

What gas is formed by this reaction? *Carbon dioxide*

Where did the gas come from? *From the acid*

Write an equation for the reaction between zinc and hydrochloric acid.



- (E) Place a small amount of sodium carbonate (Na_2CO_3) in a test tube. Add 1 to 2 cc. of sulfuric acid and observe the formation of a gas.

What gas was produced by the reaction? *Carbon dioxide*

Repeat this experiment using sodium bicarbonate (NaHCO_3) and hydrochloric acid.

What is the gas that is formed? *Carbon dioxide*

Write an equation for this reaction. *$\text{HCl} + \text{NaHCO}_3 \rightarrow \text{NaCl} + \text{H}_2\text{O} + \text{CO}_2$*

Many remedies for so called excess gastric acidity contain sodium bicarbonate.

How would the administration of this compound reduce the

¹ Dissolve 1 gm. of phenolphthalein in 100 cc. of 95 per cent alcohol.

amount of gastric acidity (HCl)? It would convert the acid + base to salt + water.

BASES

Experiment 35. Preparation of Calcium Hydroxide: Add a small piece of calcium oxide (CaO) to a test tube containing ~~10 cc.~~ of water. Warm gently until bubbles start forming in the tube. Remove from the flame and allow the tube to stand until all action has ceased. Moisten a piece of red litmus paper with the solution.

What happens? It turns blue.

Write the equation for the reaction of calcium oxide and water $\text{CaO} + \text{H}_2\text{O} \rightarrow \text{Ca(OH)}_2$

Experiment 36. Properties of Bases: (A) Add 10 cc. of water to the first two of 3 test tubes in a rack. In the first put 2 to 3 drops of a 10 per cent solution of sodium hydroxide (NaOH); in the second put 2 to 3 drops of a 10 per cent solution of ammonium hydroxide (NH_4OH); and in the third put about 10 cc. of a saturated limewater (Ca(OH)_2) solution.

Mix and taste each solution. ~~_____~~

bitter

(B) Try the effect of these solutions on red and on blue litmus paper. It turns blue red.

(C) Add a drop of phenolphthalein to each tube.

Color? purple

(D) Rub a drop of each of the more concentrated solutions of the bases (NaOH, NH_4OH , and Ca(OH)_2) between the index finger and the thumb. Rinse your fingers with water between each trial.

How do bases "feel"? Slippery

What do bases have in common to explain their similar be-

havior? (Compare their formulas) They are conta
the hydrazal ion

From the results of the above experiments fill in the following table:

	TASTE	COLOR WITH LITMUS	COLOR WITH PHENOLPHTHALEIN
Acid	<u>sour</u>	<u>blue to red</u>	<u>no reaction</u>
Base	<u>bitter</u>	<u>purple</u>	<u>turns bright red</u>

Experiment 37. Neutralization: Place 5 cc. of a 10 per cent hydrochloric acid solution in an Erlenmeyer flask. Add about 10 to 15 cc. of water and a drop of phenolphthalein. Using a pipette or a dropper add a 10 per cent solution of sodium hydroxide until the indicator changes color (about the same amount of NaOH as HCl will be needed). Swirl the flask constantly during the addition of base and do not add more than 1 drop beyond that needed to change the color. If you accidentally add too much, add a few drops of acid to change the color back and again cautiously add base. When a single drop of base changes the color, the base added has just neutralized the acid, and there is no excess of either. Taste the solution.

What substance is present? acid

Write the equation for the reaction between hydrochloric acid and sodium hydroxide. $HCl + NaOH \rightarrow$

$H_2O + NaCl$

Whenever an acid and a base neutralize each other a
salt and a water are formed.

What type of substances would be of value in the treatment of burns caused by an acid? bases

By a base? acid

SALTS

Experiment 38. Formation of Insoluble Salts: The formation of a soluble salt, sodium chloride, has been demonstrated in Experi-

ment 37. Insoluble salts may be formed as precipitates by the reaction of an acid and another salt or of two salts.

(A) To about 5 cc. of a silver nitrate (AgNO_3) ^{11 cc} solution, add a few cc.'s of hydrochloric acid.

What is the insoluble precipitate? AgCl

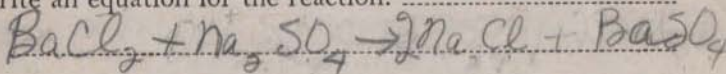
Write an equation for the reaction indicating the insoluble salt by an arrow pointing downward to the right of the

formula for the salt. $\text{HCl} + \text{AgNO}_3 \rightarrow \text{AgCl} \downarrow + \text{HNO}_3$

(B) Add a few cc.'s of a solution of sodium sulfate (Na_2SO_4) to 5 cc. of a barium chloride (BaCl_2) solution.

What is the precipitate that is formed? Barium chloride

Write an equation for the reaction.



Experiment 39. Hydrolysis of Salts: Test the reaction of solutions of the following salts to litmus and record as acid, alkaline or neutral.

Sodium carbonate

alkaline

Sodium borate (borax)

alkaline

Sodium chloride

neutral

Sodium sulfate

neutral

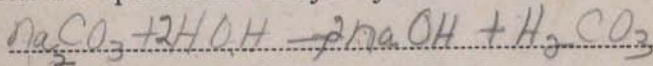
Ammonium chloride

acid

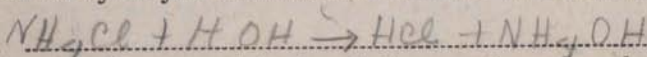
Copper sulfate

acid

Write the equation for the hydrolysis of sodium carbonate.



For the hydrolysis of ammonium chloride.



How would you explain the alkaline reaction of a solution

of sodium carbonate? It reacts to give
strong base + weak acid

Why is a sodium chloride solution neutral to litmus?

Because it was formed from
a strong base + a strong acid

Explain the acid reaction of a solution of ammonium

chloride. It reacts to give a weak
base and a strong acid

IONIZATION

SPECIAL REQUIREMENTS: Conductivity apparatus similar to that shown in Figure 34, distilled water, 1 per cent solutions of nitric, hydrochloric, acetic, boric, and sulfuric acids, 1 per cent solutions of sodium chloride and sucrose, alcohol, sodium chloride, concentrated sulfuric acid, 5 per cent solutions of the compounds listed in Experiment 41, Bogen's mixed indicator (footnote p. 50), saliva, artificial gastric juice (footnote p. 50) and pancreatic juice (footnote p. 50), urine, lemon juice, 4 per cent solution of boric acid, 0.4 per cent solutions of hydrochloric acid and sodium hydroxide, 2 per cent solutions of ammonium hydroxide and sodium bicarbonate.

Experiment 40. Electrolysis: Test the conductivity of the following solutions using an apparatus similar to that shown in Figure 34. Record your results, and an estimation of the relative intensity of the flow of the electric current through each solution, in the table.

SOLUTION	CONDUCTIVITY	INTENSITY
(A) Electrolytes vs. non-electrolytes		
Distilled water	no	
Dilute nitric acid	yes	high
Sodium hydroxide	yes	high
Sodium chloride	yes	high
Alcohol	no	
Sucrose	no	
(B) Strong vs. weak electrolytes		
Dilute hydrochloric acid	yes	high
Dilute acetic acid	yes	low
Boric acid	yes	very low
(C) Water is essential for ionization		
Dry sodium chloride	no	
Dilute sodium chloride	yes	high
Concentrated sulfuric acid	yes	high
Dilute sulfuric acid	yes	high

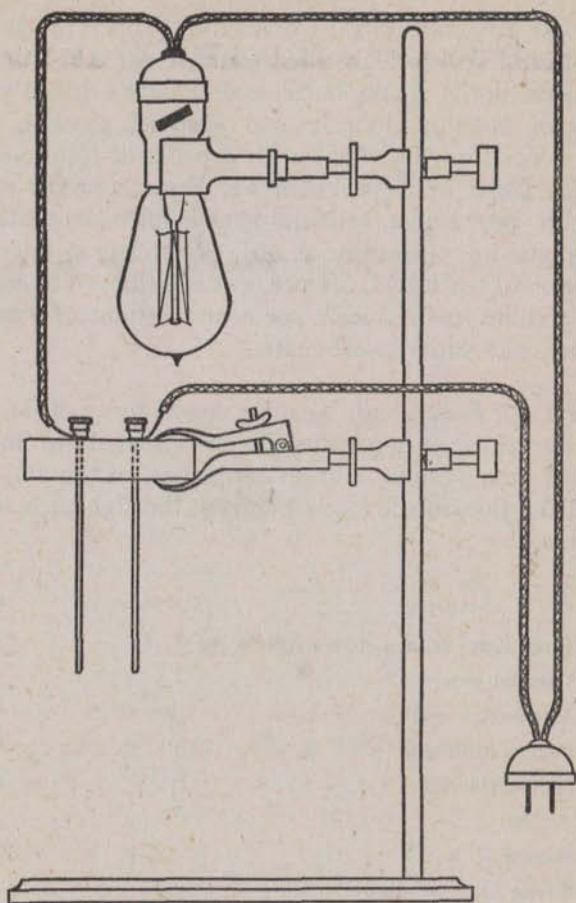


Fig. 34.—Apparatus for testing the conductivity of a solution.

Give an example of an electrolyte. *NaCl*

A non-electrolyte. *distilled H_2O*

How would you define an electrolyte? *is a sol. where H_2O solution conduct an electrical current*

A non-electrolyte? is a cgd. whose H_2O solution will not conduct an electric current

Give an example of a strong acid. Hcl

A weak acid acetic

What is the essential difference between a strong and a

weak acid? Strong acid is made up principally of ions.

Is water essential to the process of ionization? yes

When a concentrated acid is diluted with water does the

ionization of the acid increase or decrease? alts

ionization increases

What ion is responsible for the strength of acids?

hydrogen

Of bases? hydroxide

Experiment 41. The Color of Ions: Examine solutions of the following compounds. By comparison and elimination determine the color of the individual ions.

Compound	Color	POSITIVE ION		NEGATIVE ION	
		Formula	Color	Formula	Color
NaCl	colorless	Na ⁺	green	Cl ⁻	colorless
FeCl ₃	yellow	Fe	yellow	Cl ₃	colorless
NiCl ₂	green	Ni	colorless	Cl ₂	"
CoCl ₂	pink	Co	pink	Cl ₂	"
K ₂ SO ₄	colorless	K ₂	colorless	SO ₄	"
CuSO ₄	blue	Cu	blue	SO ₄	"
KMnO ₄	purple	KMn	purple	O ₄	"
K ₂ Cr ₂ O ₇	yellow	K ₂ Cr ₂	yellow	O ₇	"

What are the main differences between atoms and ions?

Their taste, color, and solubility are different.

Give an example of an element whose atom has a distinctly different color than its ion. *Copper*

Color of atom? *Copper colored*

Color of ion? *blue*

Experiment 42. Determination of pH or Hydrogen Ion Concentration: Using Bogen's mixed indicator¹ (commercial pH indicator paper may be substituted if desired), determine the pH of the following solutions:

Saliva *pH 7*

Gastric juice² *pH 1.8*

Pancreatic juice³ *pH 7.8*

Urine

Lemon juice *pH 2.6*

Boric acid *pH 3*

Dilute hydrochloric acid *pH 1.4*

Dilute sodium hydroxide *pH 1*

Ammonium hydroxide *pH 7.8*

Sodium bicarbonate *pH 7.8*

¹ Prepared by dissolving the following indicators in 500 cc. of alcohol:

0.5 gm. thymol blue

0.3 gm. dimethyl aminoazobenzene

0.2 gm. methyl red

0.4 gm. bromthymol blue

0.1 gm. phenolphthalein

Adjust the pH of the final solution to 6.0 by adding 0.1 N sodium hydroxide solution until the red color disappears, and the solution becomes yellow.

² Artificial gastric juice for pH measurements may be prepared by dissolving egg albumin (1 per cent) in a 0.4 per cent hydrochloric acid solution.

³ For this experiment artificial pancreatic juice may be prepared from egg albumin (1 per cent) in 0.5 per cent sodium carbonate solution.

Why does a solution of a weak acid (boric acid) give a pH value closer to pH 7 than a solution of a strong acid (hydrochloric acid)?

Because the more dilute the solution the lower the pH.

From the results of this experiment would you conclude that ammonium hydroxide or sodium hydroxide was the stronger of the two bases?

sodium hydroxide

Why?

What is the normal value for the pH of the blood?

7.8

What is the pH of pure distilled water?

7

Why is a solution with a pH of 7 called a neutral solution?

Because it is the turning point on the pH scale

ORGANIC COMPOUNDS

SPECIAL REQUIREMENTS: Cane sugar, chloroform, cotton-seed oil, carbon tetrachloride, methyl alcohol, ethyl alcohol, glycerol, iodine crystals, 10 per cent solution of sodium hydroxide, copper wire, 40 per cent solution of formaldehyde (formalin), Benedict's solution (footnote p. 61) concentrated sulfuric acid, 10 per cent solution of acetic acid, 1 per cent solution of butyric acid, butter, 10 per cent solution of hydrochloric acid, mixture of 1 part of sodium benzoate and 2 parts soda lime, benzene, salicylic acid, quinine sulfate tablets, chlorine water (footnote p. 58), 10 per cent ammonium hydroxide solution, strychnine sulfate tablets, potassium dichromate crystals.

ELEMENTARY COMPOSITION

Experiment 43. Test for Carbon and Hydrogen in Organic Compounds: Heat a small amount of cane sugar in a dry test tube. Hold the tube in an inclined position to avoid heating the upper portion.

What condenses in the upper part of the test tube?

Water

Continue heating until no further change is observed.

The charring of the compound indicates the presence of what element? Carbon

What element is indicated by the formation of water?

Hydrogen

What two main products are formed, besides energy, when organic compounds are completely oxidized?

Oxygen and Hydrogen

HYDROCARBONS

Experiment 44. Halogen Derivatives of Hydrocarbons: Place 1-2 cc. of chloroform in a test tube.

Describe its odor

It has a sharp strong odor

Add a few cc.'s of water to the chloroform and observe which liquid forms the lower layer in the tube.

Is chloroform heavier than water? yes

Is it soluble in water? No

To 2-3 cc. of chloroform in another test tube add a few drops of cottonseed oil. Shake the mixture.

Does the oil dissolve in the chloroform? No

Gently warm 1 cc. of chloroform in an evaporating dish and attempt to set fire to the vapors or to the chloroform with a match.

Does the chloroform burn? No

What advantage has chloroform over ether when used as an anesthetic? Chloroform doesn't burn

What disadvantage? Chloroform is more expensive

Repeat these tests using carbon tetrachloride instead of chloroform.

Describe its odor. sweet

Is it soluble in water? No

Does it dissolve the oil? _____

Will its vapors burn or support combustion? yes

Why is carbon tetrachloride used in dry cleaning? Because

it is inexpensive

ALCOHOLS

Experiment 45. Properties of Alcohols: Examine a portion (5 cc.) of methyl alcohol and record its properties as follows:

Physical state? liquid

Color? clear

Odor? sweet

Solubility in water? yesInflammability? yesColor with litmus? no reaction

Repeat these observations with ethyl alcohol and glycerol, tasting a drop of each liquid. Note the properties of ethyl alcohol.

Physical state? liquidColor? clearOdor? sharpTaste? sourSolubility in water? yesInflammability? yesColor with litmus? no reaction

Describe the properties of glycerol.

Physical state? thick liquidColor? yellowOdor? sweetTaste? sweetSolubility in water? noInflammability? noColor with litmus? no color changes

What characteristic group is common to all alcohols?

OH combined with carbon

Why do aqueous solutions of alcohols fail to turn red litmus

blue? Because it is slightly
acid.

Experiment 46. A Test for Ethyl Alcohol: Add a few crystals of iodine to 2 cc. of ethyl alcohol. After the iodine dissolves, add sodium hydroxide solution a drop at a time until the color changes to a deep yellow. Iodoform crystals form on standing and may be detected by their characteristic odor.

Describe the odor. smells like iodine

Why is ethyl alcohol sometimes called grain alcohol?

Because it is made from grain

ALDEHYDES

Experiment 47. Preparation and Properties of Formaldehyde: (A) Place 3 cc. of methyl alcohol and 3 cc. of water in a test tube. After mixing, stand the tube in a beaker of cold water. Wind a piece of copper wire around a glass tube or pencil to make a spiral about 1 inch long leaving about 8 inches of the wire for a handle. Heat the spiral in a flame until it becomes red hot then plunge it into the methyl alcohol solution. Rinse off the spiral with water and repeat the heating process several times until the solution has a sharp pungent odor.

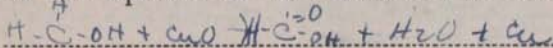
The odor is characteristic of what compound? aldehyde

When copper wire is heated, black copper oxide is formed.

Write the formula for this compound. CuO

In the above reaction the copper oxide acts as an oxidizing agent to oxidize methyl alcohol to formaldehyde.

Write the equation for the formation of formaldehyde.



(B) Place 5 cc. of Benedict's solution in each of two test tubes. To one tube, add a few drops of the formaldehyde solution prepared above and a few drops of 10 per cent sodium hydroxide solution; to the other tube, add a few drops of a 40 per cent solution of formaldehyde (formalin) and an equal number of drops of 10 per cent sodium hydroxide solution. Boil the contents of each tube for about two minutes and allow to cool.

What change do you observe in the Benedict's solution in the first tube? a red precipitate is formed

In the second tube? a red precipitate is formed

The cupric salt in Benedict's solution is reduced to insoluble cuprous oxide (Cu_2O) by compounds that contain an aldehyde group.

Aldehydes are therefore reducing agents.

What is formaldehyde used for in medicine? to test for sugar in the urine

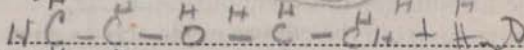
ETHERS

Experiment 48. Preparation of Diethyl Ether: Carefully add 8 to 10 drops of concentrated sulfuric acid to 3 cc. of ethyl alcohol in a test tube. Shake the tube after each addition of acid. The characteristic odor of ether should become evident.

Write the formula for diethyl ether.

Write the equation for the formation of diethyl ether from

ethyl alcohol. $\text{HC}-\text{C}(\text{OH})-\text{H} + \text{HC}-\text{C}(\text{OH})-\text{H} \longrightarrow$



What was the purpose of the concentrated sulfuric acid?

to split the water out

What is ether commonly used for in medicine? as an anesthetic

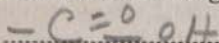
ORGANIC ACIDS

Experiment 49. Properties of Organic Acids: Examine solutions of acetic acid and butyric acid. Note the following properties of acetic acid.

Physical state? liquid
 Color? clear
 Odor? sharp
 Taste? sour
 Effect on blue litmus? turns it red

Observe the following properties of butyric acid:

Physical state? liquid
 Color? clear
 Odor? strong
 Effect on blue litmus? turns it red
 What characteristic group is common to all organic acids?



Are organic acids usually classed as strong or weak acids?

weak

Experiment 50. Test for Butyric Acid: Add 3 cc. of 10 per cent sodium hydroxide solution to a small piece of butter in a test tube. Heat gently for about a minute. Cool and acidify the mixture with 4 cc. of 10 per cent hydrochloric acid.

Upon warming the mixture the characteristic odor of what organic acid is noticeable? butyric

How does the odor compare with that of rancid butter?

Similar

Write the formula for butyric acid. $CH_3CH_2CH_2C(=O)OH$

CYCLIC COMPOUNDS

Experiment 51. Preparation and Properties of Benzene: Place a small amount of a mixture of sodium benzoate and soda lime in a test tube. Heat the tube strongly until the odor of benzene is apparent.

How does the odor compare with that of benzene in the reagent bottle? _____

Place 1-2 cc. of benzene in a test tube.

Describe its odor. like a type of gas
Add a few cc.'s of water and shake.

Is benzene soluble in water? no
To 2-3 cc. of benzene in another test tube add a few drops of cottonseed oil.

Does the benzene dissolve the fat? yes
Attempt to set fire to 1 cc. of benzene in an evaporating dish.

Does it burn? yes
Why is benzene classed as a cyclic hydrocarbon compound?
Because it is a ring hydrocarbon

Experiment 52. Preparation of an Aromatic Ester: Cautiously add about 10 to 12 drops of concentrated sulfuric acid to a mixture of 2 cc. of methyl alcohol and a small amount of salicylic acid. Warm the mixture and note the odor of the ester that is formed.

This odor is similar to that of what flavoring extract?

winter green, pepper-mint

How did the sulfuric acid function in this reaction?.....

boiled

How would you name the ester that was formed in the above reaction? sulfuric + methyl salicylic

What is this ester used for in medicine? are of wintergreen

used in medicine as a flavoring extract

ALKALOIDS

Experiment 53. Detection of Quinine and Strychnine: (A) Quinine—Dissolve one-half of a quinine sulfate tablet in 5 cc. of water. Add 1 cc. of chlorine water¹ and a few drops of ammonium hydroxide to the mixture.

¹ Add 5 cc. of concentrated hydrochloric acid to 2.5 gm. of potassium chlorate in a 500 cc. glass-stoppered bottle. Shake the mixture, then fill the bottle with distilled water. The solution should be prepared the day it is to be used.

What happens?

Now add an excess of ammonium hydroxide.

What change occurs?

(B) Strychnine: Dissolve one-half of a strychnine sulfate tablet in 5 cc. of water. To the solution add a small crystal of potassium dichromate and about 3 cc. of concentrated sulfuric acid.

Describe the color changes that occur.

Give two uses of quinine in medicine.

What is strychnine used for in the practice of medicine?

What type of cyclic compounds include the alkaloids?

CARBOHYDRATES

SPECIAL REQUIREMENTS: Glucose, fructose, sucrose, lactose, starch, Benedict's reagent (footnote p. 61), 2 per cent solutions of glucose, fructose, sucrose, and lactose, 1 per cent solution of starch, compressed yeast, small test tubes (2 to 3 in. long), iodine solution (footnote p. 63) concentrated hydrochloric acid, 10 per cent solution of sodium hydroxide.

PROPERTIES

Experiment 54. Physical Properties of Carbohydrates: Obtain small samples of the following carbohydrates: glucose, ~~fructose~~, sucrose, lactose, and starch from the reagent shelf. Taste each substance and test its solubility in water. If a carbohydrate is insoluble in cold water test its solubility in hot water by heating the mixture to boiling in a test tube.

CARBOHYDRATE	SOLUBILITY IN WATER	RELATIVE SWEETNESS
Monosaccharides		
Glucose	soluble	3
Fructose		1
Disaccharides		
Sucrose	soluble	2
Lactose	soluble	4
Polysaccharides		
Starch	not soluble	5

What kind of a solution is formed when a mixture of starch and water is boiled? colloidal

How does the starch molecule compare in size with those of the other carbohydrates? They are larger and

remain suspended.

REACTIONS

Experiment 55. Benedict's Test for Reducing Sugars: Place approximately 5 cc. of Benedict's reagent¹ in each of 5 clean test tubes, estimating the volume from the capacity of the tubes. Carefully heat the solution in one of the tubes to boiling. It should remain clear; if it does, add 8 drops of glucose solution and continue heating for a few minutes.

Result?

It turned greenish yellow

Repeat with 4 other tubes, using respectively ~~fructose~~, sucrose, lactose, and starch solutions instead of glucose.

Which sugars reduced the reagent?

Lactose, and glucose

Which did not?

sucrose and starch

What active group of the carbohydrates is the reducing group?

aldehyde or Ketone group

What has happened to this group in the carbohydrates

which do not reduce Benedict's reagent?

The aldehyde or Ketone group has been destroyed

Experiment 56. Fermentation: (Each student should choose one of the five carbohydrates in this experiment and compare results with the results of others.) Using a mortar and pestle, gently rub up a piece of compressed yeast (about the size of a pea) with 10 to 15 cc. of one of the following sugar solutions: glucose, fructose, sucrose, lactose or starch. Pour about 5 cc. of the suspension into an ordinary-size test tube. Choose a small test tube (2-3 inches long) that will readily slip into the larger test tube and fill it to overflowing with the same suspension. Tear a piece of filter paper to just fit over the mouth of the

¹ Dissolve 173 gm. of sodium citrate crystals and 100 gm. of anhydrous sodium carbonate in approximately 800 cc. of distilled water. Filter the resulting solution and add slowly, with stirring, 17.3 gm. of copper sulfate dissolved in 100 cc. of distilled water. Dilute the solution to a total volume of 1000 cc.

small tube, lay it over the opening, and turning the covered tube upside down, drop it into the large test tube (Figure 35). Set the fermentation tube in a beaker of warm water (30° – 50° C.).

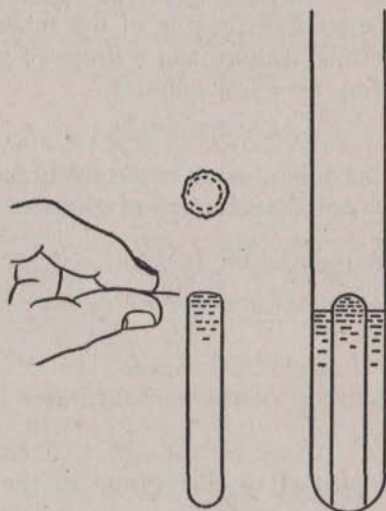


Fig. 35.—Preparation of a fermentation tube.

Gas should collect in some of the tubes before the end of the laboratory period.

What is the gas?

Carbon dioxide

What other compound is produced by fermentation?

alcohol

Which of the carbohydrates fermented? (Compare results

with others)

glucose, fructose, sucrose, and maltose

Which did not?

galactose, lactose, starch, dextrin, glycogen, and cellulose

Experiment 57. Iodine Test: Add a few drops of iodine solution¹ to about 2 cc. of starch solution in a test tube.

What happens? turns purple

Repeat the iodine test on 2 cc. portions of glucose, fructose, sucrose and lactose solutions.

Results? dark orange

When an iodine solution is spilled on a white laboratory coat or on a white uniform, what color is the spot?

blue

Why? Because of the starch present

Experiment 58. Hydrolysis of Sucrose and Starch: (A) Sucrose: To 10 cc. of sucrose solution in a small beaker add 3 drops of concentrated hydrochloric acid and boil gently for two or three minutes. Cool, and add a sodium hydroxide solution a drop at a time until the mixture is just alkaline to litmus. Test the mixture for reducing sugars with Benedict's reagent.

Results? slightly blue

Does sucrose reduce Benedict's reagent? no

What sugars are formed when sucrose is hydrolyzed?

fructose and glucose

(B) Starch: Repeat the above test using 10 cc. of starch solution instead of the sucrose solution.

Does the resultant mixture reduce Benedict's reagent?

no

Does starch reduce Benedict's reagent? no

What products may be formed when starch is hydrolyzed?

maltose, erythro dextrin, glucose

¹ Dissolve 10 gm. of potassium iodide in about 250 cc. of water. To this solution add 5 gm. of iodine, shake until dissolved, then dilute the resulting solution to 500 cc.

LIPIDS

SPECIAL REQUIREMENTS: Cottonseed oil, ether, chloroform, potassium bisulfate crystals, glycerol, palmitic acid, stearic acid, oleic acid, Hanus iodine solution (footnote p. 65), alcohol, 50 per cent sodium hydroxide solution, 25 per cent solution of sodium chloride, 5 per cent calcium chloride solution, 10 per cent solution of hydrochloric acid, cholesterol crystals, acetic anhydride, concentrated sulfuric acid.

FATS

Experiment 59. Solubility: Place 5 drops of cottonseed oil in each of 3 dry test tubes. To the first add 5 cc. of water, to the second 5 cc. of ether (Caution: highly inflammable) and to the third 5 cc. of chloroform. Mix well.

Does the fat dissolve in the water? No

In the ether? yes

✓ In the chloroform? yes

Why are such liquids as ether, chloroform, and carbon tetrachloride often called organic solvents or fat sol-

vents? Organic cpts. are soluble in organic solvents

What is the difference between an oil and a solid fat? Oil is usually unsaturated and a solid fat is saturated

Oil is usually unsaturated and a solid fat is saturated

GLYCEROL

Experiment 60. The Acrolein Test for Glycerol: Add a few crystals of potassium bisulfate (KHSO_4) to 2 drops of glycerol in a dry test tube. Heat carefully and note the irritating odor of acrolein, which is formed by the dehydration of the glycerol. Repeat the test using a fat instead of glycerol.

Results? The KHSO_4 dissolved producing an irritating odor

Have you ever noticed this odor when meat was being

fried in the kitchen? yes

Where did the acrolein come from? The dehydration of glycerol.

Why could the acrolein test be used as a general test for all fats?

Because all fats yield glycerol when heated.

FATTY ACIDS

Experiment 61. Properties of Saturated and Unsaturated Fatty Acids: Into 3 test tubes containing 5 cc. of chloroform add respectively a small amount of ^{satur.} palmitic, ^{unsatur.} stearic, and oleic acids. After the fatty acids are dissolved, add 10 drops of Hanus iodine solution¹ to each tube and note the relative speed of the disappearance of the color.

Which tubes remained colored? *oleic*

What becomes of the iodine? *In the satur. sol. it remains colored. In unsat. it added to.*

How does this test distinguish between saturated and unsaturated fatty acids? *unsaturated fatty acids*

the iodine adds on to the molecule.

SAPONIFICATION

Experiment 62. The Preparation of Soap: In a clean dry evaporating dish put 6 cc. of cottonseed oil, 5 cc. of alcohol, and 3 cc. of concentrated sodium hydroxide solution (Caution: NaOH is very caustic). Heat the mixture gently with constant stirring until the alcohol evaporates and the mass solidifies. Add 10–15 cc. of water to the contents of the dish and warm to dissolve the soap. (Save this soap solution for use in Experiment 63.)

How would you describe a soap, chemically? *Is a sodium salt of a fatty acid.*

What two products result from the saponification of a fat?

glycerol and a soap

¹ Dissolve 6.6 gm. iodine in 500 cc. of glacial acetic acid, then add 1.5 cc. of bromine to the iodine solution.

Experiment 63. The Properties of Soap: (A) Salting out of Soap: To 5 cc. of the soap solution in a small beaker add 10 cc. of a concentrated solution of sodium chloride with stirring.

What happens? *Soap comes out of solution*

Let the mixture stand a few minutes and then remove the cake of soap that has formed on the top of the liquid.

Wash the cake with cold water and attempt to form suds by shaking a piece of the soap with distilled water.

Results? *Soap forms a suds under shaking in distilled H_2O*

Is soap soluble in a salt solution? *no*

Why is this process called salting out? *Because the soap ~~salt~~ came out of solution*

(B) Formation of Insoluble Soaps: To 1 cc. of the soap solution, in a test tube, add 5 cc. of water and a few drops of a calcium chloride ($CaCl_2$) solution.

What do you observe? *A precipitate is formed*

What is the chemical nature of the precipitate? *it is*

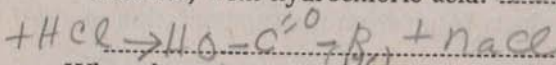
a calcium salt of a fatty acid

What is the curd that is formed when soap is used in hard water? *Calcium salt*

(C) Formation of Fatty Acids: To 5 cc. of the soap solution, in a test tube, add a hydrochloric acid solution a drop at a time until a precipitate forms.

What is the precipitate? *The fatty acid*

Write the equation for the reaction of the soap, sodium stearate, with hydrochloric acid. *$NaC_{17}H_{35}O_2 + HCl \rightarrow C_{17}H_{35}O_2 + NaCl$*



What three fatty acids are most commonly found in fats?

palmitic, oleic, stearic

$C_{16}H_{32}O_2$

$C_{18}H_{36}O_2$

C

STEROLS

omit

Experiment 64. The Properties of Cholesterol: Dissolve a few crystals of cholesterol in 2 cc. of chloroform in a clean, dry test tube. Add 10 drops of acetic anhydride and 1-3 drops of concentrated sulfuric acid and mix. Note the color changes during the first few minutes.

How would you describe them?

.....
What is a sterol?

Name two important compounds that contain the sterol nucleus.

.....

Bar

PROTEINS

SPECIAL REQUIREMENTS: Powdered egg albumin, casein, and gelatin, 2 per cent egg albumin solution, 10 per cent sodium hydroxide solution, 0.5 per cent solution of copper sulfate, 5 per cent lead acetate solution, Millon's reagent (footnote p. 69), glyoxylic acid solution (footnote p. 70), concentrated sulfuric acid, 1 per cent solutions of casein and gelatin, alcohol, 10 per cent solution of tannic acid (tannin), saturated solution of picric acid, 5 per cent solution of mercuric chloride, 2 per cent solution of silver nitrate.

TYPES OF PROTEINS

Experiment 65. Properties of Simple, Conjugated and Derived Proteins: Obtain small amounts of finely ground egg albumin, casein, and gelatin and test their solubility in cold water. If a protein is insoluble in cold water test its solubility in dilute sodium hydroxide solution; or if it is insoluble in both reagents try to dissolve it in hot water. Record the results in the following table:

	SOLUBLE IN		
	Cold Water	NaOH	Hot Water
Simple—Egg albumin	no	no	yes
Conjugated—Casein	no	no	yes
Derived—Gelatin	-	-	yes no

Define a simple protein? Simple proteins are those

that yield only amino acids or their derivatives on hydrolysis.

A conjugated protein? are simple proteins combined with some non-protein compound.

A derived protein? are protein derivatives that are produced by the action of heat, enzymes

Give an example of a protein that is insoluble in all three of the above reagents. gelatin

COLOR TESTS FOR PROTEINS

Experiment 66. Biuret Test: To 3 cc. of an egg albumin solution add an equal volume of a 10 per cent sodium hydroxide solution, mix, and add a few drops of a very dilute solution of copper sulfate (CuSO_4).

What color is produced? blue purple

Repeat the test using first a solution of casein and then one of gelatin.

Results? formed a gelatin. Turned purple
None of the three are soluble in water

Will all proteins give a positive biuret test? yes

Why?

If a protein is completely hydrolyzed will the hydrolysate give a positive biuret test? No

Explain. There has to be at least peptide linkages
before it will give a positive reaction

Experiment 67. Unoxidized Sulfur Test: Boil gently for a few minutes a mixture of 2 cc. of egg albumin solution, 5 cc. of sodium hydroxide, and a drop or two of lead acetate solution, in a small Erlenmeyer flask.

What happens? A black precipitate is formed

What is the nature of the black compound that is formed?

Copper sulfide

Where did the sulfur come from? From the amino acid

Give an example of a sulfur-containing amino acid.

Cysteine acid

Experiment 68. Millon's Test: Add 5-10 drops of Millon's reagent¹ to 3 cc. of an egg albumin solution in a test tube, and heat gently over a flame. The mercury salts in the reagent first precipitate the protein which gradually changes color on heating.

¹ Dissolve 100 gm. of mercury in 150 cc. of concentrated nitric acid by gently heating the mixture under a hood. Dilute the resulting solution with 2 volumes of distilled water.

What color is produced? red

What amino acid present in proteins is responsible for this reaction? tryptophan

Experiment 69. Hopkins Cole Test: Mix 2 cc. of egg albumin solution and 3 cc. of a glyoxylic acid solution¹ in a test tube. Pour this solution carefully down the side of a test tube containing 3 cc. of concentrated sulfuric acid (Caution: a strong acid) so that the two solutions do not mix. A distinct color should develop at the junction of the two liquids. If the color does not appear within a few minutes, tap the tube to produce agitation at the zone of contact.

Results? A purple color was formed between

What amino acid in proteins responds to this test? tryptophan

PRECIPITATION OF PROTEINS

Experiment 70. Heat Coagulation: Fill a test tube about two-thirds full of egg albumin solution and heat the upper portion of the solution.

Results? The protein coagulates + forms precipitate

Repeat the test on a solution of casein and on a solution of gelatin.

Results? _____

How would you detect the presence of albumin in a pathological urine specimen? _____

Experiment 71. Alcohol Precipitation: Add about 5 cc. of alcohol (95 per cent) to 3 cc. of egg albumin solution.

Results? _____

¹ To 10 gm. of powdered magnesium in a large Erlenmeyer flask, add distilled water until the magnesium layer is well covered. Slowly pour 250 cc. of a cold saturated solution of oxalic acid into the flask, shaking continuously and cooling the contents in a stream of running water. Filter, then add 25 cc. of glacial acetic acid to the filtrate, and dilute to 1 liter with distilled water.

Why are alcoholic solutions used to disinfect areas of the skin before surgery?

Experiment 72. Precipitation by Alkaloidal Reagents: To about 5 cc. of egg albumin solution in a test tube add 1-2 cc. of tannic acid solution.

What happens?

Repeat the experiment using picric acid solution instead of tannic acid.

Results? *A gray looking color with a dark precipitate*

Why are these two reagents effective in the treatment of burns? *Because they precipitate the proteins*

forms a layer over burn to prevent air to enter or H₂O to leave

Experiment 73. Precipitation by Salts of Heavy Metals: Add a mercuric chloride solution a drop at a time to a solution of egg albumin.

Does a precipitate form?

Repeat the experiment with a solution of silver nitrate.

Results?

Of what advantage is the administration of egg white or milk to patients who have taken mercuric chloride or silver nitrate internally? *Causes coagulation of*

proteins

Why must this treatment be followed by an emetic? *In order to get rid of the*

metals

ENZYMES AND DIGESTION

SPECIAL REQUIREMENTS: 1 per cent solution of pepsin, 10 per cent sodium hydroxide solution, 0.5 per cent solution of copper sulfate, 1 per cent starch solution, iodine solution (footnote p. 63), Benedict's reagent (footnote p. 61), whole milk, saturated litmus solution, 2 per cent solution of steapsin, fibrin, 1 per cent solution of pepsin in 0.4 per cent hydrochloric acid solution, 2 per cent trypsin solution in 0.5 per cent sodium carbonate solution, fresh 1 per cent solution of rennin (footnote p. 74), pepsin solutions for Experiment 79: 1 per cent pepsin in 0.4 per cent hydrochloric acid, and 0.5 per cent sodium carbonate solution, pancreatin solutions for Experiment 79: 2 per cent pancreatin in 0.5 per cent sodium carbonate solution, and in 0.4 per cent hydrochloric acid solution, boiled solutions of these enzymes (footnote p. 75), fresh bile, concentrated nitric acid, powdered sulfur, gallstones, ether, chloroform, acetic anhydride, concentrated sulfuric acid.

ENZYMES

Experiment 74. Chemical Nature of Enzymes: To 3 cc. of a solution of pepsin add an equal volume of 10 per cent sodium hydroxide solution, mix, and add a few drops of a very dilute solution of copper sulfate.

Results?

A positive Biuret test indicates that enzymes are what type

of compounds?

DIGESTION OF CARBOHYDRATES, FATS, AND PROTEINS

Experiment 75. Digestion of Starch: Collect about 10 cc. of saliva for use in this experiment and add 2 cc. of the saliva to 10 cc. of 1 per cent starch solution (cooked) in a test tube. Place the tube in a beaker of warm water (approximately 40° C.). At one minute intervals remove a drop of the digestion mixture and add it to a drop of iodine solution. (A spot plate is very convenient.)

What color is formed with the iodine at the beginning of the experiment?

Describe the color changes that occur as digestion proceeds.

.....
When a drop of the solution fails to give a color with iodine, apply Benedict's test to the contents of the tube. (5 cc. of reagent plus 8 drops of solution, heat.)

Is there a reduction?

What is the compound, formed by the digestion of starch by salivary amylase, that is responsible for a positive

Benedict's test?

What are the intermediate products of digestion called?

.....
What is the active enzyme of saliva?

Experiment 76. Digestion of Fats: In each of two marked test tubes place 5 cc. of milk and 1 cc. of saturated litmus solution. To the first tube add also 1 cc. of a solution of steapsin, and to the second add an equal quantity of distilled water. Both tubes should be blue after the above additions. Let the tubes stand for about an hour in warm water (approximately 40° C.).

Do you observe any change in color in either tube?

.....
Which tube?

What acid product has been formed to cause the color change of litmus?

Where has the acid come from?

Experiment 77. Digestion of Proteins: Place small pieces of fibrin, of approximately the same size, in each of three marked test tubes. Add 5 cc. of an acid solution of pepsin to the first tube, 5 cc. of an alkaline solution of trypsin to the second, and an equal

quantity of distilled water to the third. Place the tubes in a beaker of warm water (approximately $40^{\circ}\text{C}.$) for an hour. Examine the contents. As the fibrin is digested, it becomes soluble and the pieces become progressively smaller.

Has digestion of the protein occurred in the first tube?

.....

The second?

The third?

What soluble products are formed when an insoluble protein is digested by enzymes?

What products result from the complete digestion of a protein?

The third tube is called a control.

Why did digestion fail to occur in this tube?

.....

Why is a control tube important in enzyme experiments?

.....

Experiment 78. Clotting of Milk: To 5 cc. of milk add 5 drops of a freshly prepared solution of rennin.¹ Place in warm water (approximately $40^{\circ}\text{C}.$) for 20 minutes. Examine.

What has happened?

.....

What is the function of rennin in the above reactions?

.....

Calcium ions are necessary for the precipitation of paracasein. .

Where did they come from in this experiment?

.....

¹ Mix 1 gm. of reasonably fresh, commercial rennin or rennet with 100 cc. of water. Stir until dissolved.

Would rennin activity be more important in the stomach of an infant or in that of an adult?

Why?

FACTORS THAT INFLUENCE ENZYME ACTION

Experiment 79. Effect of pH on Enzyme Action: Into each of 6 marked test tubes put a piece of fibrin (approximately the same size pieces); then to the tubes add 5 cc. of solution as follows:

Tube No.	5 cc. of
1	Acid solution of pepsin
2	Alkaline solution of pepsin
3	Acid solution of pepsin, boiled ¹
4	Alkaline solution of pancreatin
5	Acid solution of pancreatin
6	Alkaline solution of pancreatin, boiled ¹

Place the tubes in a beaker of warm water (40° C.) for an hour. Examine.

In which tubes has digestion occurred?

Does pepsin require an acid or an alkaline reaction for its optimum activity?

What is the protein-splitting enzyme of pancreatin?

Does it act best in an acid or an alkaline solution?

What is meant by "optimum pH"?

Experiment 80. Effect of Temperature on Enzyme Action: Into each of 3 test tubes place 10 cc. of 1 per cent starch solution (cooked) and 2 cc. of saliva. Boil the contents of one tube for at

¹ To insure the destruction of enzyme activity by boiling, the pepsin and pancreatin solutions should be placed in a beaker and boiled for 5 to 10 minutes. Any loss of volume by evaporation should be corrected by the addition of distilled water.

76 Enzymes and Digestion

least 3 minutes and then place it and a second tube in a beaker of warm water. (Approximately 40° C.). Place the third tube in a beaker of ice water. At one minute intervals take a drop from the second tube and test with iodine.

How long does it take before a drop fails to give the starch-iodine color? minutes.

Now apply the iodine test to a drop from each of the other tubes.

Result?

What effect does boiling have on an enzyme solution?

.....
How does a low temperature affect the activity of an enzyme?

What is meant by "optimum temperature"?

BILE

Experiment 81. Bile Pigments: Filter 3 cc. of fresh bile through the cone of a filter paper. Carefully add a drop of concentrated nitric acid into the cone of the paper and observe the various colors that form as the bile pigments are oxidized.

Describe the color changes.

.....
From what substances are the bile pigments formed in the body?

What bile pigments are responsible for the color of the urine?

The feces?

Experiment 82. Reduction of Surface Tension by Bile Salts: Place 1 cc. of bile and 5 cc. of water in a clean test tube. Place an equal volume of water in a second tube. Holding the tubes on a level

with the eyes, sprinkle a little powdered sulfur upon the surface of the two liquids.

Results?

What are the two major types of bile salts?

.....
In cases of jaundice, bile is present in the blood stream.

What effect do bile salts have on the red blood cells?

.....
.....
Experiment 83. Gallstones: Grind a small gallstone in a dry mortar with 10 cc. of ether (Caution, highly inflammable). Filter through a small filter paper into a dry evaporating dish. Rinse the material in the mortar and on the filter paper with a few cc.'s of ether. Set the beaker aside until the ether evaporates. Dissolve the residue in 3 cc. of chloroform and transfer to a clean dry test tube. Add 10 drops of acetic anhydride and 1 to 3 drops of concentrated sulfuric acid, mix.

Note the color changes during the first few minutes.

.....
A positive test indicates the presence of what constituent in gallstones? (See Experiment 64.)

What other substances may be present in a gallstone?

.....

NORMAL URINE

SPECIAL REQUIREMENTS: Bottles for urine collection, toluene, graduated cylinders 1000 cc. and 2000 cc., urinometers, phenol red indicator (footnote p. 80), 1 per cent acetic acid solution, fresh 1 per cent urease solution (footnote p. 80), 10 per cent sodium carbonate solution, Benedict's uric acid reagent (footnote p. 81), concentrated hydrochloric acid, saturated solution of picric acid, 10 per cent sodium hydroxide solution, 10 per cent solution of nitric acid, 2 per cent silver nitrate solution, ammonium molybdate solution (footnote p. 82) 10 per cent hydrochloric acid solution, 5 per cent solution of barium chloride.

COLLECTION OF A 24-HOUR SPECIMEN

Since many of the general properties of urine and all of the quantitative determinations are based on a 24-hour urine specimen it may be helpful to give detailed directions for its collection.

Obtain a 2 to 3 liter bottle and place in it 10 cc. of toluene as a preservative. A 24-hour sample refers to the urine produced by the kidneys in that time; it is therefore necessary that the bladder be empty at the beginning and at the end of the collection period. If it is desired to start the collection at 8 A.M. the bladder should be emptied at exactly that time and the urine discarded. All subsequent urine for 24 hours should be placed in the bottle. After each addition of urine the bottle should be shaken to mix the urine with the toluene. The sample should be kept in a cool place. At exactly 8 A.M. the following morning the bladder should be emptied again, this time adding the urine to that in the bottle. The sample should be collected so that the end of the collection period comes a short time before the urine is examined in the laboratory.

GENERAL PROPERTIES

Experiment 84. General Properties of Urine: Choose three representative 24-hour specimens for this experiment, preferably one with a high volume, one with a normal and another with a low volume.

(A) Volume: Measure the volume of the three 24-hour samples in a large graduated cylinder, recording the results in the table below.

(B) Specific Gravity: By means of a urinometer (Figure 36) determine the specific gravity of the three samples. It is best to adjust the temperature of the urine to the temperature indicated on the urinometer, which is the temperature at which the scale was calibrated. The urinometer scale should be read at the point where it touches the surface of the urine. There should be no foam on the surface of the urine and the urinometer should not touch the sides of the cylinder when readings are taken. Record the results in the table.

(C) Color: Describe the color of the three specimens, in the following table.

(D) Reaction: Test the reaction of the three specimens to litmus paper, recording your observations in the table.

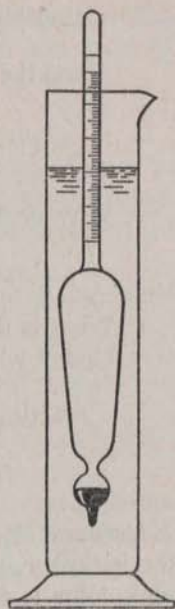


Fig. 36.—A
urinometer.

SPECIMEN No.	VOLUME cc.	SPECIFIC GRAVITY gm. / cc.	COLOR	REACTION TO LITMUS
1				
2				
3				

What volume of urine is normally excreted in 24 hours?

Would this volume tend to increase or decrease in hot weather?

After prolonged exercise?

In cold weather?

From the results of this experiment, what general rela-

tionships exist between the volume, the specific gravity, and the color of a 24-hour urine specimen?

Why is the color of urine directly related to the volume?

What is the average pH of normal urine?

Under what conditions would the urine have an alkaline reaction?

NORMAL CONSTITUENTS—ORGANIC

Experiment 85. Urea: To 5 cc. of fresh urine add a few drops of the indicator, phenol red.¹ A yellow solution should result. If a pink color is formed add very dilute acetic acid until the pink tint is replaced by a yellow one. Now add 2 cc. of urease solution² and note the time required for the liberation of ammonia in sufficient amount to produce a red solution. (Alkaline color of indicator.)

Results?

Write the equation for the reaction of urease on urea.

Approximately how many grams of urea are excreted per

day on a normal diet?

How could you alter the quantity of urea excreted in 24 hours?

¹ Grind 0.1 gm. of powdered phenol red in a mortar with 28.2 cc. of 0.01 N sodium hydroxide solution. Rinse the mixture into a flask with distilled water and dilute to a volume of 250 cc.

² Prepare a 1 per cent solution of urease by dissolving urease powder or tablets in distilled water. The solution should be prepared the same day it is used.

Experiment 86. Uric Acid: To 3 cc. of urine in a test tube add 2 cc. of a 10 per cent sodium carbonate solution and 1 cc. of Benedict's uric acid reagent.¹

Results?

Add 1 cc. of concentrated hydrochloric acid to 50 cc. of urine in a beaker, stir well and set aside until the next laboratory period. Examine the characteristic pigmented crystals that have formed on the sides of the beaker.

Why are uric acid crystals not normally found in the urine?

What dietary constituents give rise to uric acid in the urine?

Experiment 87. Creatinine: To 5 cc. of urine in a test tube add 1 cc. of saturated picric acid solution. Make the solution alkaline by adding 1 cc. of 10 per cent sodium hydroxide solution and observe the color.

Results?

Why is the quantity of creatinine excreted in 24 hours relatively constant for a normal individual?

What is the creatinine coefficient?

Under what conditions may the creatinine excretion be increased?

Decreased?

¹ Dissolve 100 gm. pure sodium tungstate in about 600 cc. of water. Add 50 gm. of pure arsenic pentoxide, followed by 25 cc. of 85 per cent phosphoric acid and 20 cc. of concentrated hydrochloric acid. Boil the mixture for 20 minutes, cool and dilute to 1 liter with distilled water.

NORMAL CONSTITUENTS—INORGANIC

Experiment 88. Chlorides: Acidify 10 cc. of urine with 1 cc. of dilute nitric acid solution and add a few drops of a silver nitrate solution.

Results?

What is the principal source of chlorides in the diet?

Experiment 89. Phosphates: Acidify 10 cc. of urine with 1 cc. of dilute nitric acid solution and add 2 cc. of ammonium molybdate solution.¹ Warm the solution, allow the tube to stand a few minutes and observe the color.

Results?

What type of diet would increase the daily excretion of phosphates?

Experiment 90. Sulfates: Place 10 cc. of urine in a test tube, acidify with 1 cc. of dilute hydrochloric acid, heat to boiling and add a few drops of a barium chloride solution.

Results?

Write the equation for this reaction.

What is the principal source of sulfates in the diet?

¹ Dissolve 25 gm. of ammonium molybdate in 5 N sulfuric acid solution and dilute to 1 liter with the same acid solution.

PATHOLOGICAL URINE

SPECIAL REQUIREMENTS: Benedict's solution (footnote p. 61), urine specimens containing approximately 0.2, 0.5, and 1.0 per cent glucose, normal urine, urine specimens containing approximately 0.2, 0.5 and 1.0 per cent albumin, 10 per cent solution of acetic acid, fresh saturated solution of benzidine in glacial acetic acid, 3 per cent solution of hydrogen peroxide, urine containing blood (footnote p. 84), powdered sulfur, urine containing bile (footnote p. 84), special ammonium sulfate reagent (footnote p. 85), sodium nitroprusside reagent (footnote p. 85), urine containing acetone bodies (footnote p. 85).

Experiment 91. Glucose: Apply Benedict's test to normal urine and to three samples of diabetic urine of varying degree of severity. Heat 5 cc. of Benedict's solution to boiling, add 8 drops of urine and continue to heat for 2 to 3 minutes. Compare the extent of reaction in each tube. Results:

Normal

Slight diabetic

Moderate diabetic

Severe diabetic

If the conditions, i.e. volume and heating time, are uniform in all four tests, accurate comparable results may be obtained.

Is there any reducing sugar normally present in urine?

.....
Under what conditions might glucose be excreted in the
urine?

Experiment 92. Albumin: Apply the heat coagulation test to normal urine and to three specimens of urine containing varying amounts of albumin. Fill a test tube about two-thirds full of urine and heat the upper portion to boiling. Compare with the cooler portion. Add a few drops of dilute acetic acid to dissolve

any phosphates which may have precipitated. Compare the extent of coagulation in each tube. Results:

Normal

Slight albuminuria

Moderate albuminuria

Severe albuminuria

Uniform conditions of heating will insure comparable results.

What pathological conditions result in the excretion of albumin in the urine?

Why is albumin not normally present in the urine?

Experiment 93. Blood: Place 2 cc. of urine in a test tube; add 3 cc. of a fresh solution of benzidine, and mix thoroughly. Add 1 cc. of hydrogen peroxide and note the color produced. Apply this test to normal urine and to a sample of urine containing blood.¹ Results:

Normal urine

Pathological urine

What abnormal condition results in the presence of blood in the urine?

What is hemoglobinuria?

Experiment 94. Bile: Sprinkle a small amount of powdered sulfur on the surface of a sample of normal urine and a sample of urine containing bile.² Results:

Normal urine

Pathological urine

¹ Add 1 cc. of blood to 1 liter of urine.

² Add 50 cc. of bile to 950 cc. of urine.

What other simple test may be used to detect the presence of bile in the urine?

What is jaundice and how is it usually caused?

Experiment 95. Acetone Bodies: Add 5 cc. of the special ammonium sulfate reagent¹ to 5 cc. of urine in a test tube, and mix. Then add 1 cc. of a nitroprusside reagent,² shake, and let stand a minute. Observe the color that is formed. Apply this test to normal urine and to urine containing acetone bodies.³ Results:

Normal urine

Pathological urine

Name several conditions that would result in the excretion of acetone bodies in the urine?

Experiment 96. Unknown Pathological Urines: Obtain two unknown specimens of pathological urine from the instructor. By applying the above tests, determine which pathological constituent is present in each of the unknowns.

Unknown No. 1

Unknown No. 2

¹ Mix 200 cc. of saturated ammonium sulfate solution with 200 cc. of concentrated ammonium hydroxide solution.

² Dissolve 10 gm. of sodium nitroprusside in 90 cc. of distilled water and add 1 cc. of concentrated nitric acid.

³ Add 3 cc. of acetone to 1 liter of urine.

VITAMINS

SPECIAL REQUIREMENTS: Saturated solution of trichloroacetic acid in chloroform, cod liver oil, bismuth potassium iodide reagent (footnote p. 86), thiamin solution (footnote p. 86), nicotinic acid solution (footnote p. 87), solution of phosphomolybdic acid (footnote p. 87), fresh solution of stannous chloride (footnote p. 87), 10 per cent sodium hydroxide solution, 1 per cent solution of ascorbic acid, 5 per cent ferrous sulfate solution, solution of aniline in hydrochloric acid (footnote p. 88).

Experiment 97. Vitamin A: To 3 cc. of a saturated solution of trichloroacetic acid in chloroform in a test tube add 2 drops of cod liver oil and shake.

What color is produced?

The vitamin A in cod liver is responsible for this color reaction.

What outstanding condition results from the lack of vitamin A in the diet?

What pigments are closely related to this vitamin?

Experiment 98. Thiamin (Vitamin B₁): To 5 cc. of a bismuth potassium iodide reagent¹ add 1 cc. of a thiamin solution.²

What happens?

The thiazole ring in thiamin is responsible for the formation of the colored precipitate.

What are some of the early symptoms that develop on a diet deficient in thiamin?

¹ Add 2 gm. of bismuth nitrate ($\text{Bi}(\text{NO}_3)_3$) to 500 cc. of 5 per cent potassium iodide solution. Let stand a few hours and filter the solution.

² Prepare a thiamin solution containing 0.5 to 1.0 mg. per cc. by dissolving thiamin chloride in distilled water.

Name three good sources of this vitamin in the diet.

.....

Experiment 99. Nicotinic Acid: Add 2 cc. of a nicotinic acid solution¹ to 2 cc. of a solution of phosphomolybdic acid.²

What happens?

To the precipitated nicotinic acid mixture add 1 cc. of a freshly prepared solution of stannous chloride.³

What color is produced?

What disease is caused by the lack of nicotinic acid in the diet?

Why is pellagra more prevalent in the Southern states?

.....

.....

Why is nicotinic acid sometimes called niacin?

.....

Experiment 100. Ascorbic Acid (Vitamin C): Add 2 drops of a 10 per cent solution of sodium hydroxide to 2 cc. of a 1 per cent solution of ascorbic acid in a test tube. To this slightly alkaline solution add 2 cc. of a 5 per cent solution of ferrous sulfate (FeSO_4) solution and allow the mixture to stand until a color develops.

Results?

What are some of the symptoms of subacute scurvy?

.....

.....

¹ Prepared by dissolving nicotinic acid in distilled water; should contain about 5 mg. per cc. of solution.

² Dissolve 12.5 gm. of phosphomolybdic acid in 200 cc. of 10 per cent hydrochloric acid solution.

³ Dissolve 40 gm. of stannous chloride in 100 cc. of concentrated hydrochloric acid. This solution should be prepared shortly before the period in which it is used

What common type of diet would lack sufficient quantities of ascorbic acid?

Name three good sources of this vitamin.

Experiment 101. Vitamin D: To approximately 2 cc. of cod liver oil in a test tube add 2 cc. of a solution of aniline in hydrochloric acid.¹

What color is produced?

The vitamin D in cod liver oil is responsible for this color formation.

Why are children given daily supplements of cod liver oil?

What other substances are involved with vitamin D in the formation of bone and teeth?

Why do children require less vitamin D in the summer than in the winter months?

¹ Prepared by mixing 1 part of concentrated hydrochloric acid and 15 parts of aniline.

HORMONES

SPECIAL REQUIREMENTS: 1 per cent solution of sodium acetate, adrenalin solution (1:100000), solution of mercuric chloride (1:1000), alcoholic solution of estrogenic hormones (footnote p. 89), β naphthol reagent (footnote p. 89), distilled water, alcoholic solution of androgenic hormones (footnote p. 90), 2 per cent meta-dinitrobenzene in alcohol, 5 N solution of potassium hydroxide.

Experiment 102. Adrenalin: Add 1 cc. of a 1 per cent solution of sodium acetate to 1 cc. of a dilute solution of adrenalin (1:100000) in a test tube. To this mixture add 4 to 5 drops of a 1:1000 solution of mercuric chloride and warm the contents of the tube to 50° C.

What happens?

Why is adrenalin said to be antagonistic to insulin?
.....
.....

List three important therapeutic uses of adrenalin.
.....

Experiment 103. Female Sex Hormones: Place 1 cc. of an alcoholic solution of estrogenic hormones¹ in a test tube in a boiling water bath until the alcohol evaporates. To the residue add 1 cc. of the β naphthol reagent,² shake the tube and heat it in the boiling water bath for 3 minutes, shaking twice during this period. Cool in tap water, then carefully add 1 cc. of distilled water and return the tube to the boiling water bath for 3 minutes.

What color is produced?

¹ An alcoholic solution of estrone containing from 0.03 to 0.06 mg. per cc. is satisfactory.

² Dissolve 2.5 gm. of β naphthol in 100 gm. of concentrated sulfuric acid, cooled in ice water.

What are the three main hormones of the follicle?

What are the functions of the estrogenic hormones?

Experiment 104. Male Sex Hormones: Place 5 drops of a solution of androgenic hormones¹ dissolved in 95 per cent alcohol in a test tube. Add 5 drops of a 2 per cent solution of meta-dinitrobenzene dissolved in 95 per cent alcohol and 5 drops of a 5 N potassium hydroxide solution, stopper the tube and mix well. Set the tube in your desk for 30 minutes or until a definite color develops.

What color is produced?

What two androgenic hormones are usually found in male urine?

What are the functions of the androgens?

¹ An alcoholic solution of androsterone containing from 0.05 to 0.2 mg. per cc. is satisfactory.

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